

Memorandum

To: Stephanie Vaughn, EPA Region 2

Elizabeth Buckrucker, USACE

From: Frank Tsang and Sharon Budney

Date: November 10, 2011

Subject: Toxicity Test, Bioaccumulation Split Sample Data Comparison and Comments on

the CPG Draft 2009 Bioaccumulation Tissue Chemistry Data for the Lower Passaic

River Study Area, September 19, 2011

At the request of the United State Environmental Protection Agency (USEPA) and the United States Army Corps of Engineers (USACE), CDM Federal Programs Corporation (CDM) reviewed the Draft 2009 Bioaccumulation Tissue Chemistry Data report for the Lower Passaic River Study Area, dated September 19, 2011, prepared by Windward Environmental LLC on behalf of the Cooperating Parties Group (CPG) for the Lower Passaic River (LPR) Restoration Project.

As a part of the 2009 LPR investigation the Louis Berger Group, Inc. (LBG) collected split samples of sediment, fish tissue, crab tissue, and worm tissue for laboratory analysis during the 2009 Fish and Benthic Tissue Sampling program conducted by the Cooperating Parties Group (CPG) for the LPR Remedial Investigation (RI). Split sample toxicity tests using test organisms were also conducted.

The following information has been extracted from LBG's memorandum of September 27, 2011 titled Split Sample Data Comparison 2009 Lower Passaic River Fish and Benthic Tissue Sampling Oversight, table and figure numbers have been modified from the original document to minimize confusion in their sequencing within this summary:

Samples will be referred to as CPG samples or USEPA samples for clarity. The significant bioaccumulation split sample comparison findings are summarized below.

- Worm Tissue Comparison. The worm tissue split sample comparison was constrained because
 two split sample pairs only (10% of 20 CPG samples) were generated by the oversight
 program. In cases where both the CPG laboratory and the USEPA laboratory generated
 detected results, the percent difference generally met the criteria.
- <u>Toxicity Testing</u>. The toxicity test result pairs met the percent difference criteria for organism survival except for one instance; however, all but one of the result pairs failed to meet the percent difference criteria for organism growth with the CPG results consistently higher than the USEPA laboratory results.

Oversight Program Summary

Oversight was conducted in accordance with the Final Oversight Quality Assurance Project Plan (QAPP) for Biological Sampling, Community Surveys, Toxicity and Bioaccumulation Testing prepared by Malcolm Pirnie, Inc. and Battelle (August 2009) and associated approved QAPP modifications.

The bioaccumulation split sample program consisted of:

- 2 worm tissue split samples from bioaccumulation testing
- 5 Ampelisca abdita 10-day survival toxicity tests
- 5 Chironomus dilutus 10-day survival and growth toxicity tests
- 10 Hyalella azteca 28-day survival and growth toxicity tests (5 freshwater and 5 estuarine tests)

Data Comparison Methodology

To examine the parent and split sample datasets for potential bias, CPG sample and USEPA split sample data were plotted in three different formats for selected analytical parameters:

- A line plot of absolute concentration for the paired samples. The line plot provides insight on the relative magnitudes and patterns of concentrations measured by both analytical programs for the paired samples.
- A bivariate scatter plot of the detected concentrations. The bivariate scatter plot illustrates the relationship between the CPG sample and USEPA split sample data, and in particular, highlights potential systematic bias if the points fall consistently above or below the 1:1 line.
- A line plot of percent difference. The percent difference (%D) is defined as the difference between the USEPA and CPG sample concentrations, divided by the USEPA sample concentration. Consequently, a negative %D indicates a CPG result that is higher than the USEPA result, while a positive %D indicates a CPG result that is lower than the USEPA result. This plot provides a visual indication of the extent of positive and negative differences between the two datasets. The red dashed lines on the plot correspond to 40%D and -67%D. These criteria correspond to 50% relative percent difference (RPD, the CPG's field duplicate acceptance criterion), converted to %D values. Note that RPD and %D are similar mathematical functions that allow a comparison of two values. %D is commonly used when one of the two values is known or accepted, whereas RPD is more commonly used when both values are uncertain (for example, for comparison of field duplicates).

In addition to the preparation of the above listed data comparison plots (Figures 1 through 49) the tests described below were also conducted for the CPG and USEPA data pairs where a result was obtained above the detection limit for both samples. The findings of these tests are summarized in Table 1.

- The average and standard error was calculated for the ratio of CPG result to USEPA result (result greater than 1 indicates on average that the CPG's laboratory detected higher concentrations for a particular parameter; result less than 1 indicates that on average the USEPA laboratory detected a higher concentration of a particular parameter).
- %D was compared to the criteria of 40%D and -67%D (equivalent to 50% RPD). The 50% RPD criteria are derived from the CPG's field duplicate evaluation criterion.
- The Wilcoxon Signed Rank test was used to calculate p-values. The p-value is an indicator of the presence of a bias or difference between the datasets. P-values less than 0.05 indicate a statistically significant difference between results.

Table 1 also contains the column "Overall Split Sample Comparison (Same or Different)," which presents the judgment of the data reviewers regarding the comparability of the split sample data. An opinion that the datasets were comparable (entry of "Same") was based on the following lines of evidence and associated criteria:

Average ratio of CPG to USEPA data within 0.7 to 1.3.

- %D within 40% to -67% for the majority of the sample pairs (one or two exceedances permitted if other lines of evidence indicated comparability of the CPG and USEPA data).
- No statistical bias.

Where the cells in Tables 1 contain multiple values, the second value was calculated with outliers removed from the comparison.

Worm Tissue (Bioaccumulation Testing)

The data comparison for worm tissue was constrained because the oversight program yielded two split sample pairs only (10% of 20 CPG samples, as per the planned split sampling frequency). P-values could not be calculated due to the small dataset. Where both the CPG laboratory and the USEPA laboratory provided detected results, the %D was generally within the acceptable range.

Toxicity Test Data

The split sample toxicity testing results generated by American Aquatic Testing, Inc. (USEPA laboratory) were reviewed by Battelle to evaluate the data quality (refer to attached Verification Reports). The following data verification findings were provided by Battelle:

- Ampelisca abdita (A. abdita) 10-day survival tests oversight data are acceptable without reservation.
- Chironomus dilutus (C. dilutus) 10-day survival and growth tests data are acceptable with reservations because hardness varied beyond the QAPP requirements and may have impacted the bioavailability of metals to the test organisms.
- Hyalella azteca (H. azteca) 28-day survival and growth estuarine tests data are acceptable
 with reservations due to excessive variation in alkalinity and hardness compared to the QAPP
 requirements.
- *H. azteca* 28-day survival and growth freshwater tests data are acceptable with reservations due to variation in hardness.

The comparison of the CPG and USEPA laboratory toxicity test survival and growth results is presented in Tables 2a and 2b. With the exception of one *H. azteca* test, the results pairs for mean survival met the %D criteria (see Table 2a). For the growth data, the comparison was strikingly different with all but one sample pair exceeding the %D criteria. The CPG growth data were consistently higher than the USEPA data.

Comments on the September 19, 2011 CPG Draft 2009 Bioaccumulation Tissue Chemistry Data for the Lower Passaic River Study Area are included on the attached pages.

$\frac{Comments}{Draft\ 2009\ Bioaccumulation\ Tissue\ Chemistry\ Data\ for\ the\ Lower\ Passaic\ River\ Study\ Area\ Dated\ September\ 19,2011}$

No.	Page No.	Specific Comments		
1	Page 2, First paragraph, Second sentence	Please delete "analytical data", or revise appropriately when referring to Table 1-1 as no analytical data were collected during the habitat and avian surveys.		
2	Page 3, Table 1-1	Under the Column titled "QAPP/Sampling Plan Citation" the AECOM's QAPPs for RM 10.9 and small-volume CWCM are listed as in preparation. Please revise with the correct dates as both the draft and final RM 10.9 and small-volume CWCM QAPPs have been completed.		
3	Page 13, Section 3.1 It is recommended that text be included to provide an explanation as to why a screening not run prior to the <i>N. virens</i> test initiation as noted for <i>L. variegatus</i> in Section 3.1.2.			
4	Page 15, First paragraph	Please provide a more detailed explanation as to why the 4-day screening test was conducted on <i>L. varigatus</i> . Was this driven because of concerns of toxicity associated with salinity or quality of test organisms? In addition, it is suggested that a brief discussion of test results be included other than just referencing Appendix H.		
5	Page 18, Second paragraph, second and third sentences	The text states that 66 grams of tissue was required for analysis and that all <i>N. virens</i> samples had sufficient mass; however, one <i>N. virens</i> sample weighed 63 grams. Please revise the text appropriately.		
6	Page 19, Table 3-5 The number of <i>Lumbriculus</i> samples (13/15) submitted for pesticide analysis differs from presented in Table 3 of Appendix A (12/15). Please revise accordingly.			
7	Page 46, Second paragraph, first sentence	The text states that 13 pesticides were detected in <i>N. virens</i> samples and 16 in <i>L. variegates</i> . Review of Table 4-8 indicates a total of 10 and 19, respectively. These totals do not take into account total concentrations of parent compounds and isomers. It appears that the discrepancy lies within including these values with individual compounds; however, it is still unclear how the total values of 13 and 16 were derived. Please clarify, and if needed, revise accordingly.		
8	Page 59, Section 5.8, last sentence	The sentence begins with "Nine-five percent of the samples" It seems as though the writer meant ninety-five percent Please review this statement and revise accordingly		

Table 1 - 2009 Lower Passaic River Worm Tissue Split Sample Comparison Summary Table

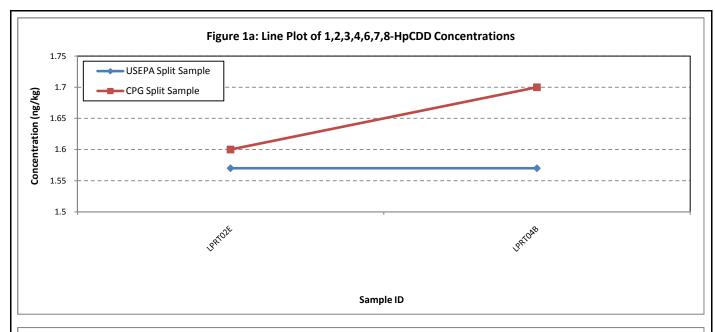
	l able .	1 - 2009 Lower Passaic River Worm	rissue Split Sampi	e Comparison Summary 18	abie			
	Number of	Number of Split Sample Pairs where	Average Ratio of	Comparison to Percent	P-Value (for	Presence of	Overall Split Sample	
Parameter	Split Sample	Detected Concentrations were	CPG to USEPA with	Difference Criteria (for	detected	Statistical Bias	Comparison (Same or	
	Pairs	Reported by USEPA and CPG	Standard Error (for	detected pairs)	pairs)	(Yes or No)	Different)	
Dioxin/Furans	Dioxin/Furans							
1,2,3,4,6,7,8-HpCDD	2	2	0.95 ± 0.029	Within Range	NA	NA	Inconclusive	
1,2,3,4,6,7,8-HpCDF	2	2	1 ± 0.033	Within Range	NA	NA	Inconclusive	
2,3,7,8-TCDD	2	2	1.1 ± 0.059	Within Range	NA	NA	Inconclusive	
2,3,7,8-TCDF	2	2	0.86 ± 0.054	Within Range	NA	NA	Inconclusive	
OCDD	2	2	1.1 ± 0.049	Within Range	NA	NA	Inconclusive	
OCDF	2	2	1.2 ± 0.12	Within Range	NA	NA	Inconclusive	
Total TCDD	2	2	1.1 ± 0.11	Within Range	NA	NA	Inconclusive	
Metals	ı		T	T	ı	T	1	
Arsenic	2	2	0.86 ± 0.082	Within Range	NA	NA	Inconclusive	
Barium	2	2	0.76 ± 0.11	Within Range	NA	NA	Inconclusive	
Cadmium	2	1	1 ± 0	Within Range	NA	NA	Inconclusive	
Chromium	2	2	1 ± 0.012	Within Range	NA	NA	Inconclusive	
Cobalt	2	2	1 ± 0.045	Within Range	NA	NA	Inconclusive	
				Outside of Range for one				
Copper	2	2	1.6 ± 0.14	samples	NA	NA	Inconclusive	
Iron	2	2	1.1 ± 0.046	Within Range	NA	NA	Inconclusive	
Lead	2	2	1.2 ± 0.29	Within Range	NA	NA	Inconclusive	
				Outside of Range for two				
Mercury	2	2	5.3 ± 0.33	samples	NA	NA	Inconclusive	
Nickel	2	2	1 ± 0.075	Within Range	NA	NA	Inconclusive	
Zinc	2	2	0.93 ± 0.00075	Within Range	NA	NA	Inconclusive	
РАН	ı		T	T	ı	T		
				Outside of Range for two				
Anthracene	2	2	0.44 ± 0.034	samples	NA	NA	Inconclusive	
Benzo[a]anthracene	2	2	1 ± 0.15	Within Range	NA	NA	Inconclusive	
Benzo[a]pyrene	2			NA	NA	NA	Inconclusive	
Chrysene	2	2	0.87 ± 0.012	Within Range	NA	NA	Inconclusive	
Fluoranthene	2	2	0.81 ± 0.043	Within Range	NA	NA	Inconclusive	
Indeno[1,2,3-cd]pyrene	2			NA	NA	NA	Inconclusive	
Naphthalene	2			NA	NA	NA	Inconclusive	
				Outside of Range for one				
Phenanthrene	2	2	0.68 ± 0.098	samples	NA	NA	Inconclusive	
Pyrene	2	2	1.1 ± 0.057	Within Range	NA	NA	Inconclusive	
Pesticides							-	
2,4'-DDD	2			NA	NA	NA	Inconclusive	
2,4'-DDE	2			NA	NA	NA	Inconclusive	
2,4'-DDT	2			NA	NA	NA	Inconclusive	
4,4'-DDD	2	2	0.99 ± 0.047	Within Range	NA	NA	Inconclusive	
4,4'-DDE	2			NA	NA	NA	Inconclusive	
4,4'-DDT	2			NA	NA	NA	Inconclusive	
Dieldrin	2	2	1.1 ± 0.045	Within Range	NA	NA	Inconclusive	
gamma-Chlordane	2	2	1.2 ± 0.18	Within Range	NA	NA	Inconclusive	
Percent Lipids								
				Outside of Range for two				
Percent Lipids (Bligh-Dyer 1959 Method)	2	2	3.2 ± 0.17	samples	NA	NA	Inconclusive	
Percent Lipids (Laboratory SOP MSU-018								
R05)	1	1	0.87 ± 0	Within Range	NA	NA	Inconclusive	
РСВ			•	<u>-</u>		•	•	
Total PCB	2	2	0.95 ± 0.0035	Within Range	NA	NA	Inconclusive	
3,3',4,4'-Tetrachlorobiphenyl (BZ 77)	2	2	0.97 ± 0.02	Within Range	NA	NA	Inconclusive	
3,4,4',5-Tetrachlorobiphenyl (BZ 81)	2	1	-	Within Range	NA	NA	Inconclusive	
2,3,3',4,4'-Pentachlorobiphenyl (BZ 105)	2	2	1 ± 0.017	Within Range	NA	NA	Inconclusive	
2,3,4,4',5-Pentachlorobiphenyl (BZ 114)	2	2	0.97 ± 0.023	Within Range	NA	NA	Inconclusive	
2,3',4,4',5-Pentachlorobiphenyl (BZ 118)	2	2	1 ± 0.036	Within Range	NA	NA	Inconclusive	
2,3',4,4',5'-Pentachlorobiphenyl (BZ 123)	2	2	1.1 ± 0.0045	Within Range	NA	NA	Inconclusive	
3,3',4,4',5-Pentachlorobiphenyl (BZ 126)	2	1		Within Range	NA	NA	Inconclusive	
2,3,3',4,4',5-Hexachlorobiphenyl +	_	_						
2,3,3',4,4',5'-Hexachlorobiphenyl (BZ								
156 + BZ 157)	2	2	1 ± 0.054	Within Range	NA	NA	Inconclusive	
2,3',4,4',5,5'-Hexachlorobiphenyl (BZ 167)	2	2	0.98 ± 0.025	Within Range	NA NA	NA NA	Inconclusive	
3,3',4,4',5,5'-Hexachlorobiphenyl (BZ 167)	2	0	0.30 ± 0.023	Within Range Within Range	NA NA	NA NA	Inconclusive	
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2,3,3',4,4',5,5'-Heptachlorobiphenyl (BZ 189)	2	2	1.1 ± 0.086	Within Range	NA	NA	Inconclusive	

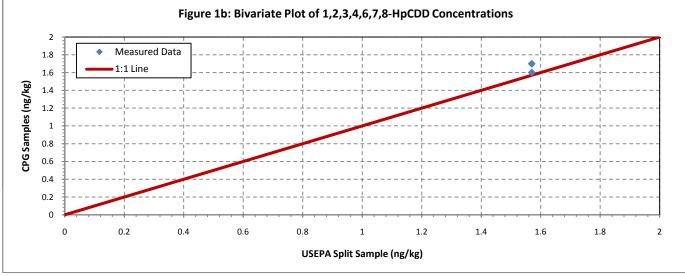
Table 2a - 2009 Lower Passaic River Toxicity Test Split Sample Comparison

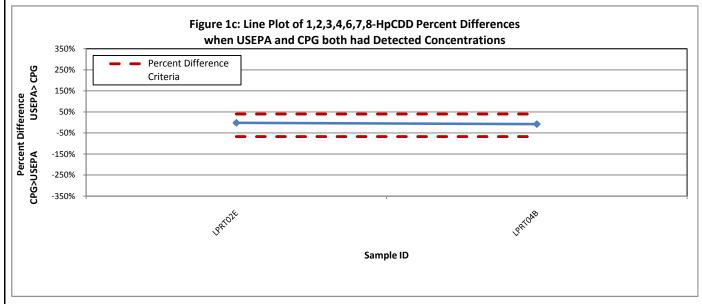
	Organism Type	Mean Perce	Relative Percent	
Sample Location ID		USEPA	CPG	Difference
LPRT01F	Ampelisca abdita	82	81	1.2
LPRT01G	Ampelisca abdita	98	92	6.3
LPRT02A	Ampelisca abdita	86	85	1.2
LPRT02F	Ampelisca abdita	74	79	6.5
LPRT03A	Ampelisca abdita	90	58	43.2
LPRT11A	Chironomus dilutus	87.5	76.3	13.7
LPRT11C	Chironomus dilutus	77.5	78.8	1.6
LPRT11D	Chironomus dilutus	93.8	83.8	11.3
LPRT11E	Chironomus dilutus	92.5	87.5	5.6
LPRT16A	Chironomus dilutus	97.5	70	32.8
LPRT01F	Hyalella azteca	85	87.5	2.9
LPRT01G	Hyalella azteca	87.5	82.5	5.9
LPRT02A	Hyalella azteca	75	80	6.5
LPRT02F	Hyalella azteca	82.5	83.8	1.5
LPRT03A	Hyalella azteca	87.5	76.3	13.7
LPRT11A	Hyalella azteca	88.8	79.4	11.2
LPRT11C	Hyalella azteca	92.5	55	50.8
LPRT11D	Hyalella azteca	70	67.5	3.6
LPRT11E	Hyalella azteca	50	76.3	41.6
LPRT16A	Hyalella azteca	58.8	66.7	12.5

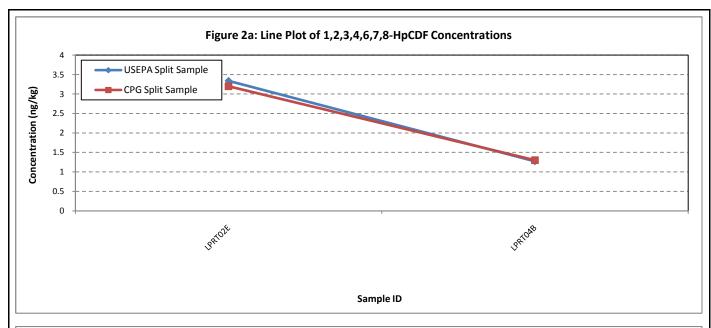
Table 2b - 2009 Lower Passaic River Toxicity Test Split Sample Comparison

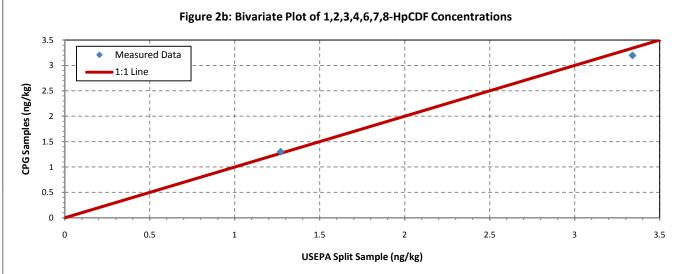
	Organism Type	Mean Growth	Weight (mg)	Relative Percent
Sample Location ID		USEPA	CPG	Difference
LPRT11A	Chironomus dilutus	0.516	1.054	68.5
LPRT11C	Chironomus dilutus	0.522	1.571	100.2
LPRT11D	Chironomus dilutus	0.528	1.047	65.9
LPRT11E	Chironomus dilutus	0.534	0.779	37.3
LPRT16A	Chironomus dilutus	0.731	1.289	55.3
LPRT01F	Hyalella azteca	0.197	0.429	74.1
LPRT01G	Hyalella azteca	0.255	0.425	50
LPRT02A	Hyalella azteca	0.206	0.373	57.8
LPRT02F	Hyalella azteca	0.204	0.375	59
LPRT03A	Hyalella azteca	0.263	0.648	84.6
LPRT11A	Hyalella azteca	0.207	0.596	96.9
LPRT11C	Hyalella azteca	0.203	0.586	97.1

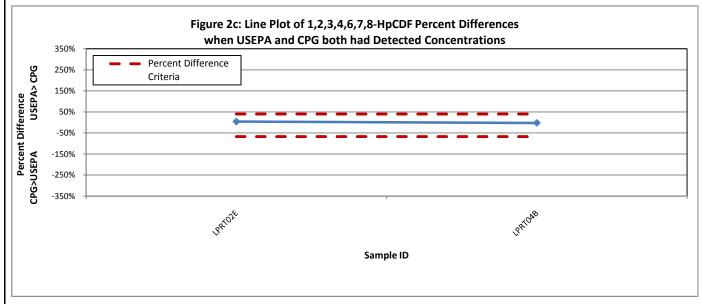




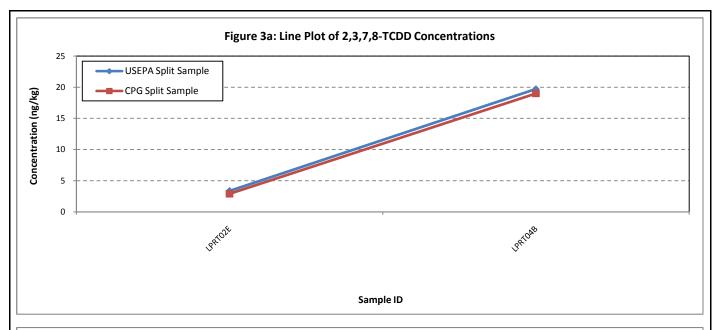


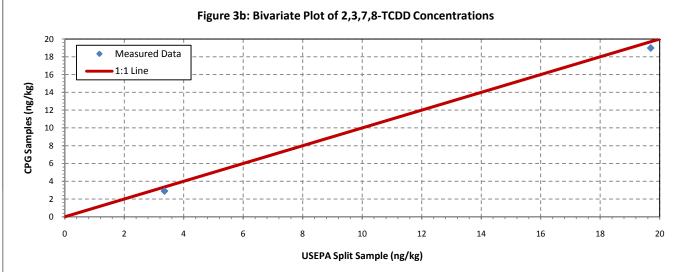


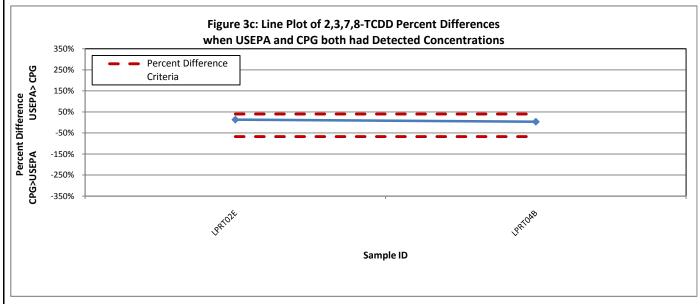




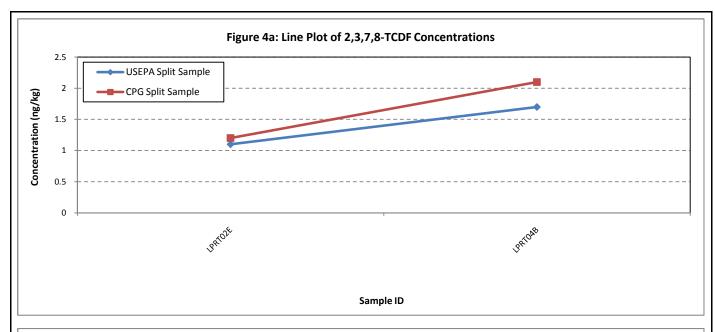


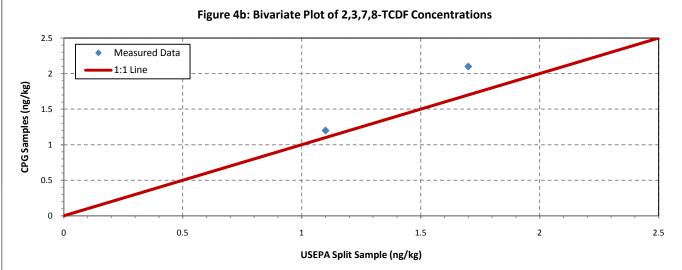


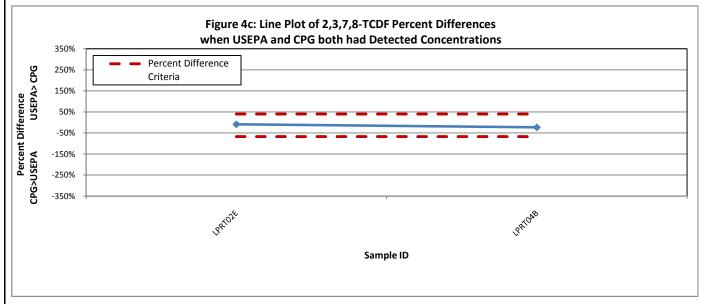




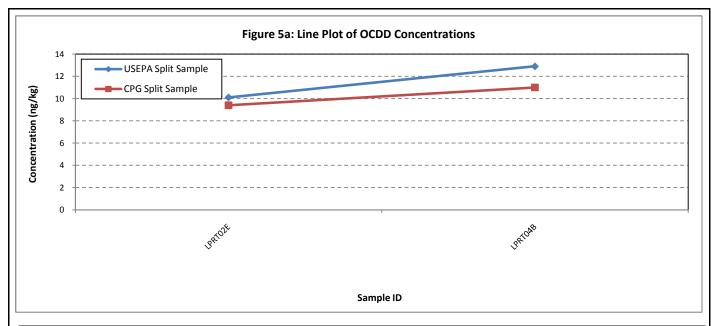


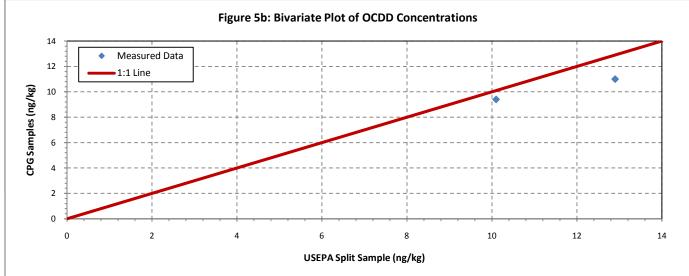


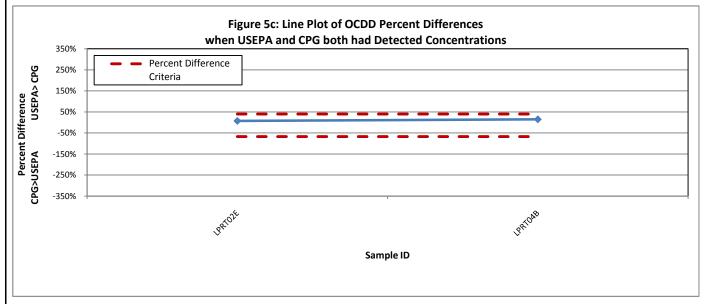


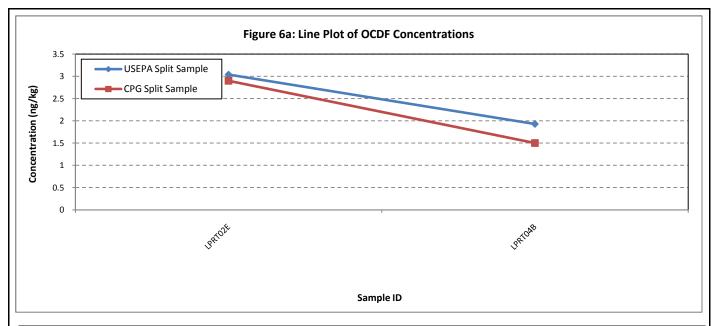


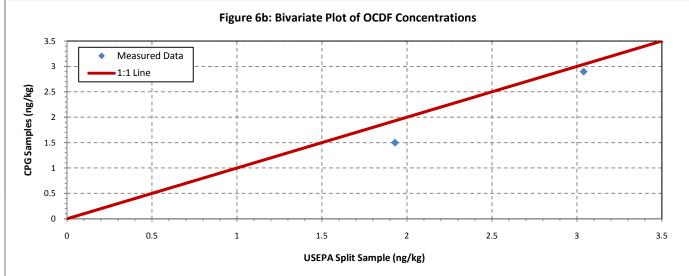


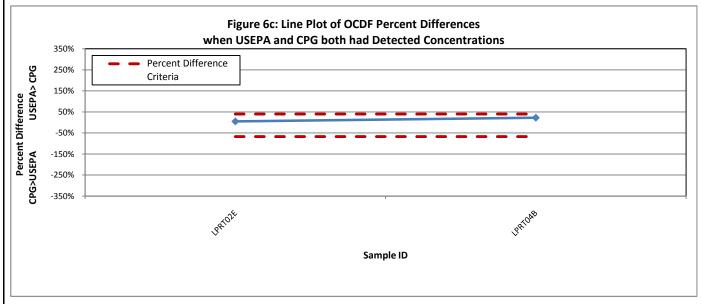


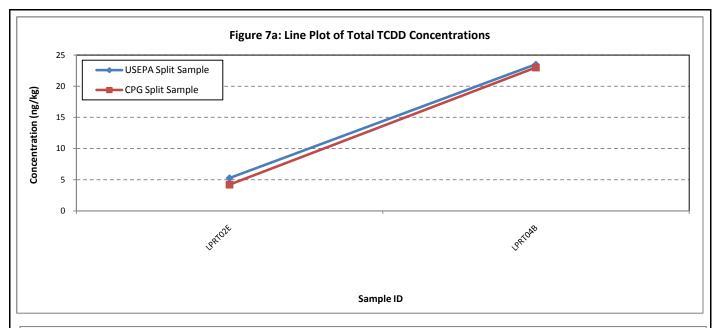


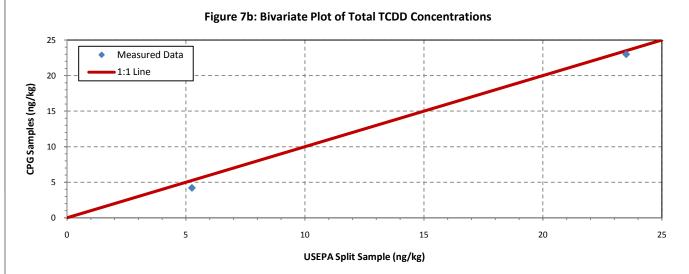


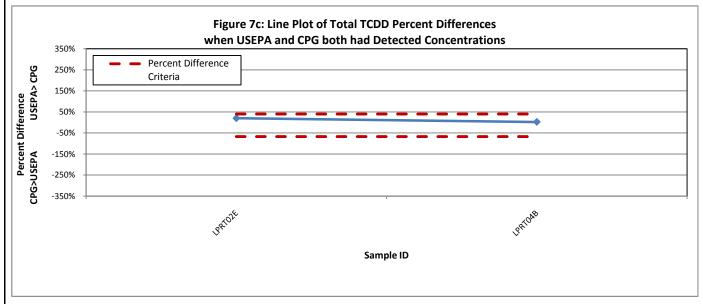


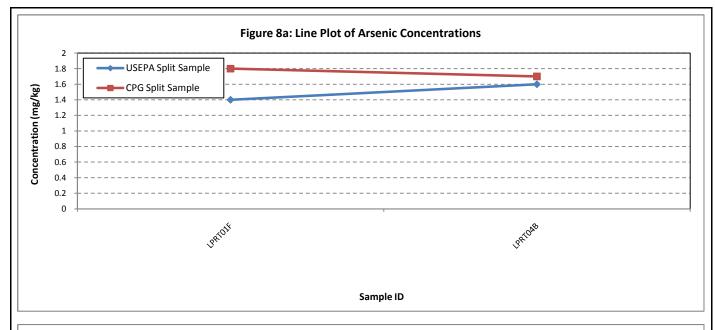


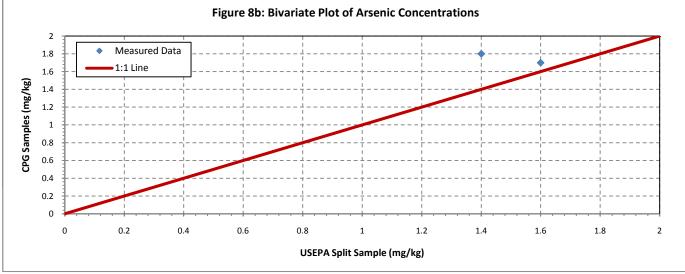


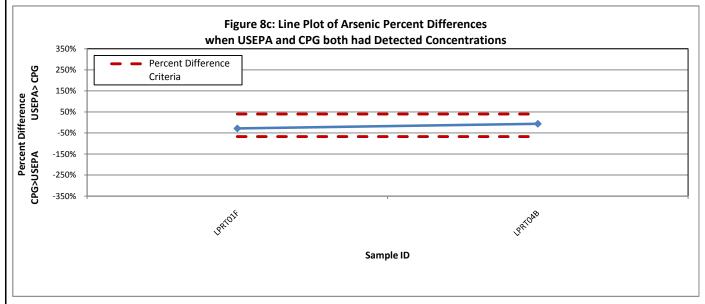


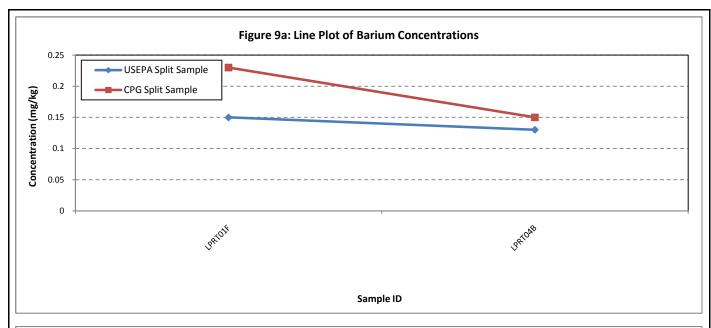


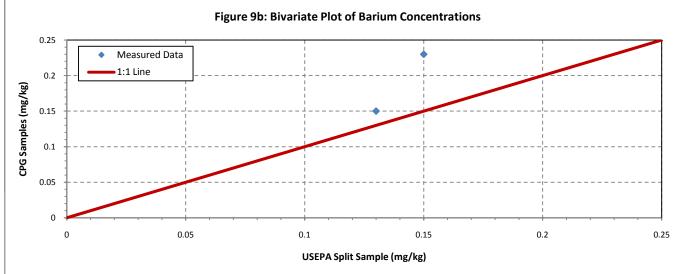


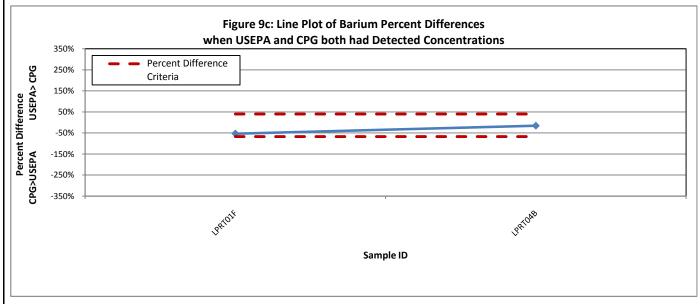


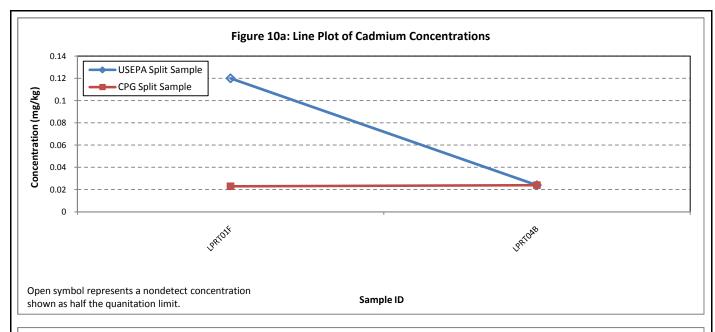


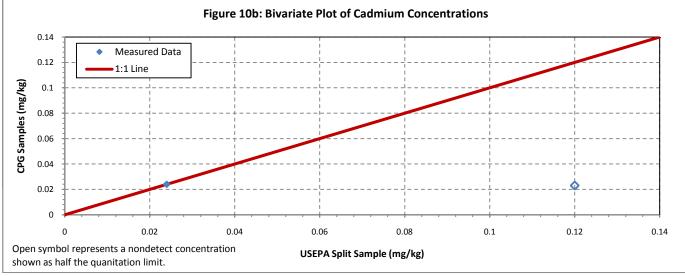


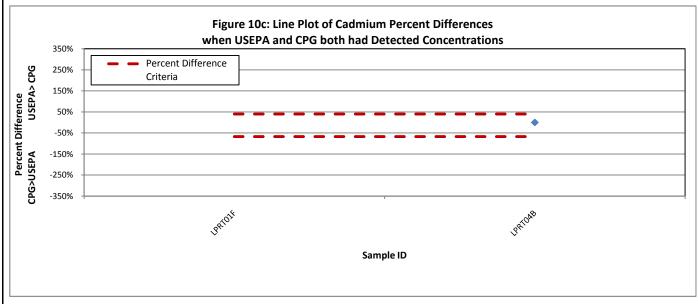




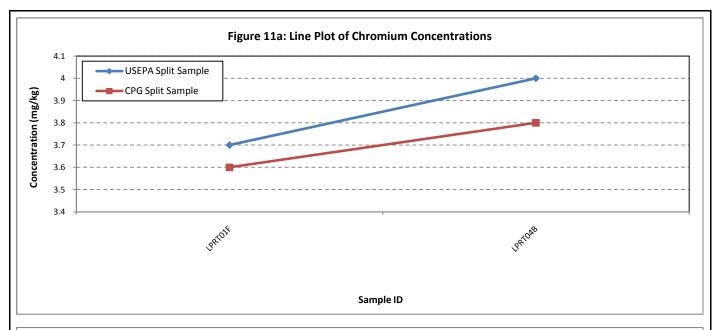


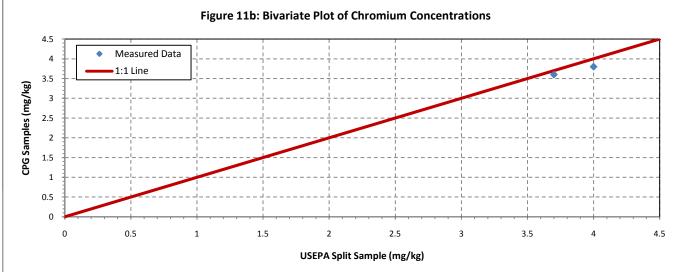


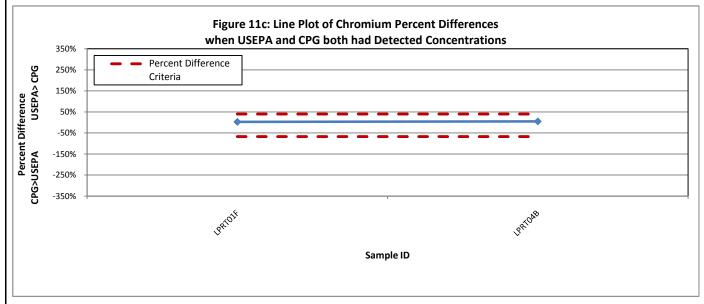


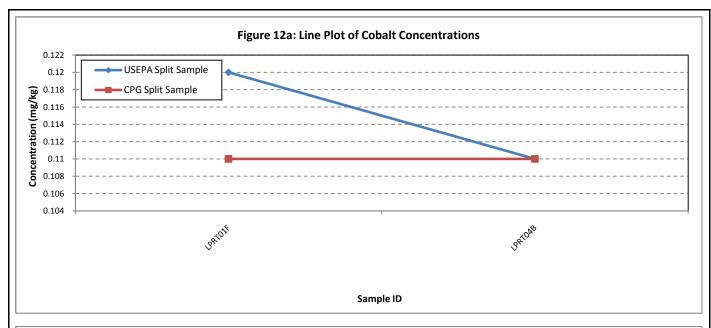


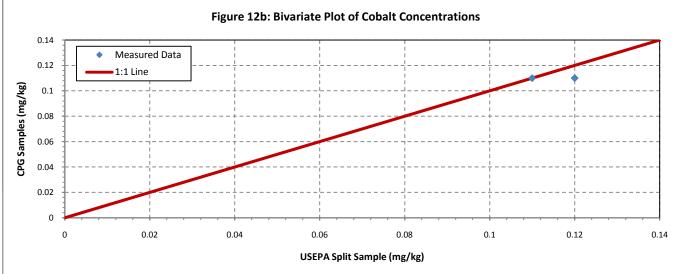


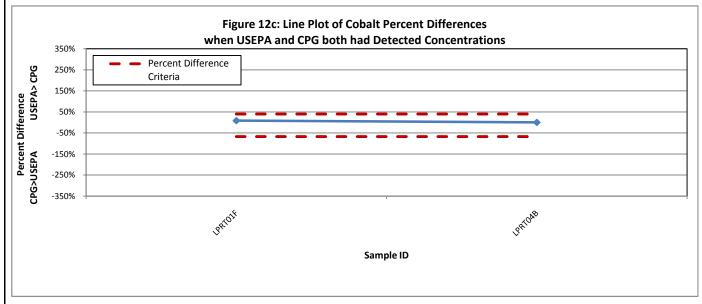


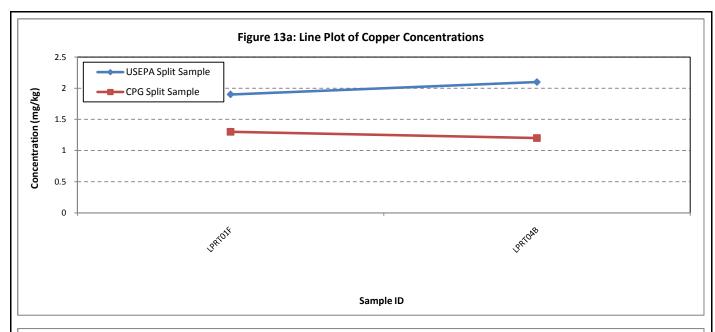


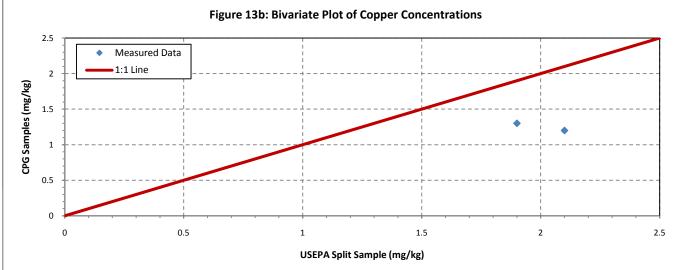


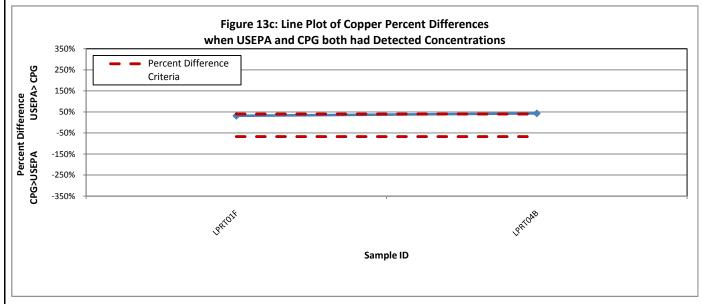


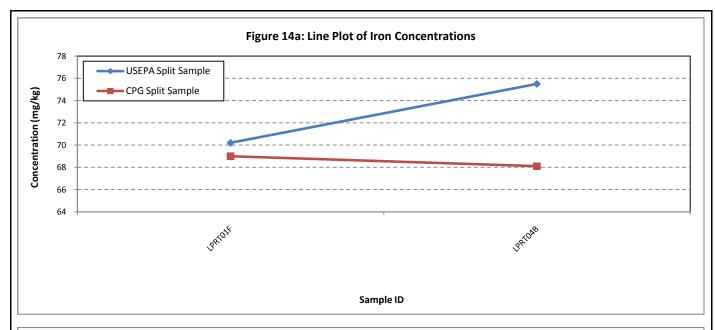


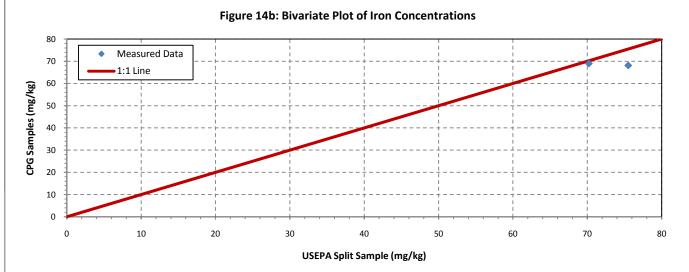


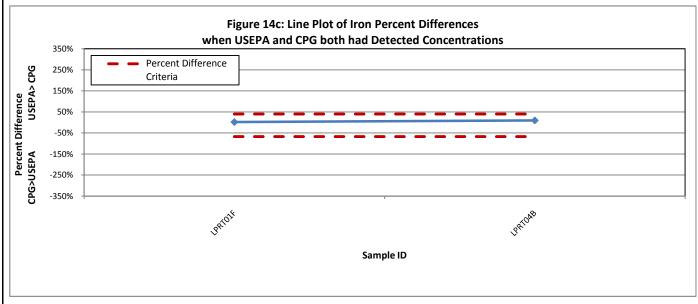




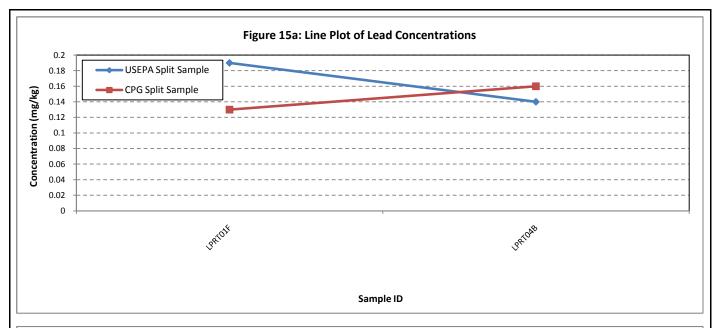


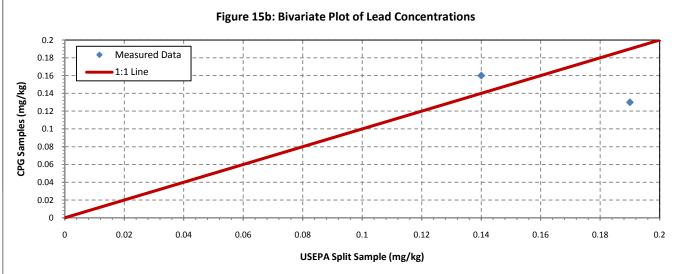


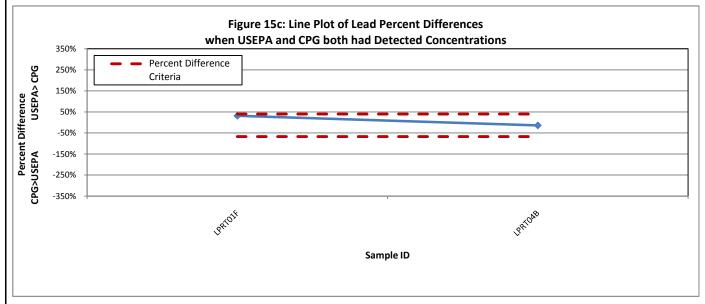




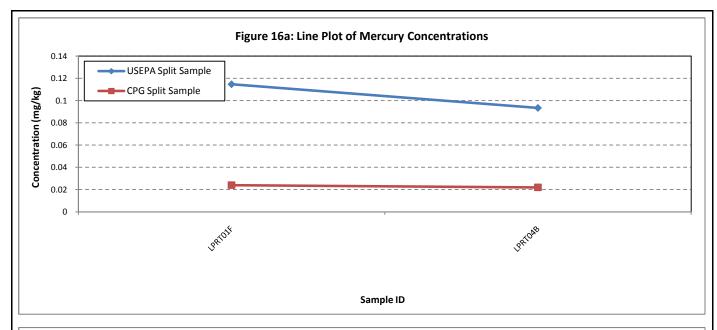


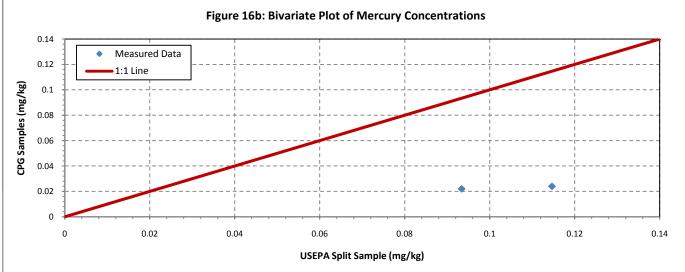


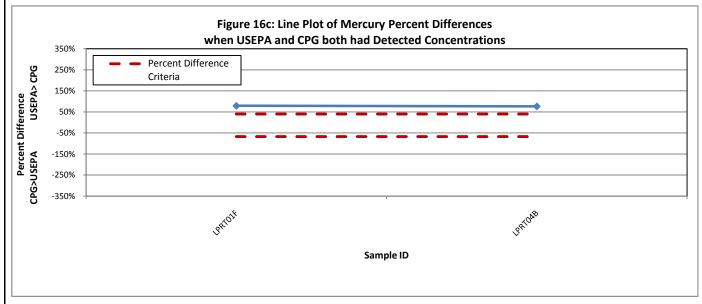




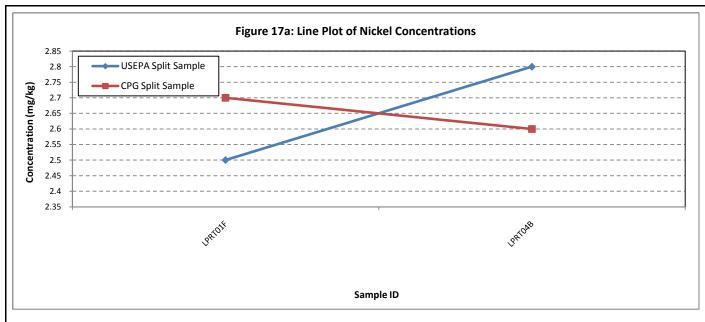


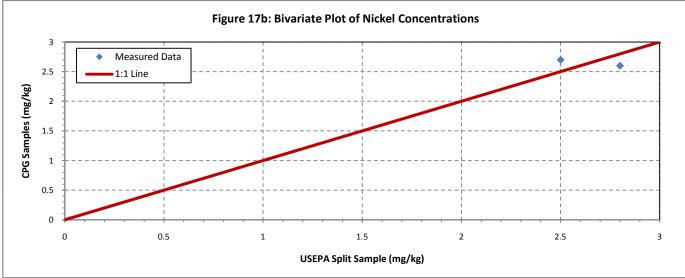


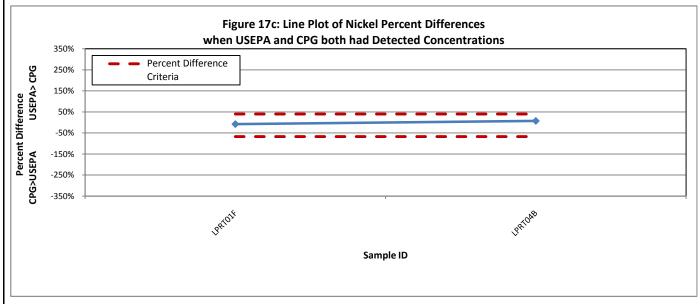


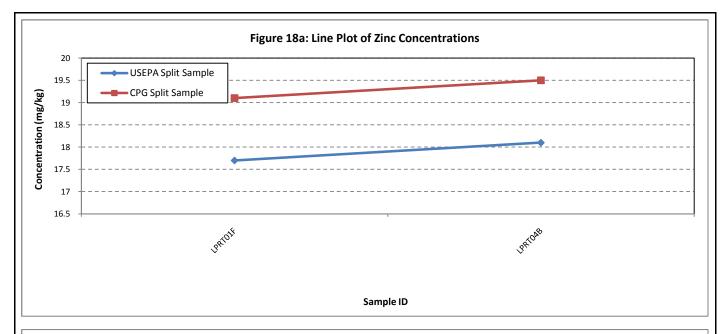


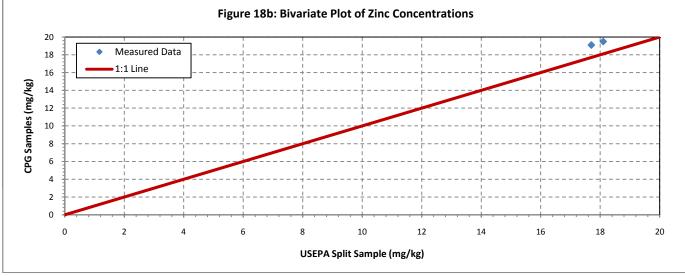


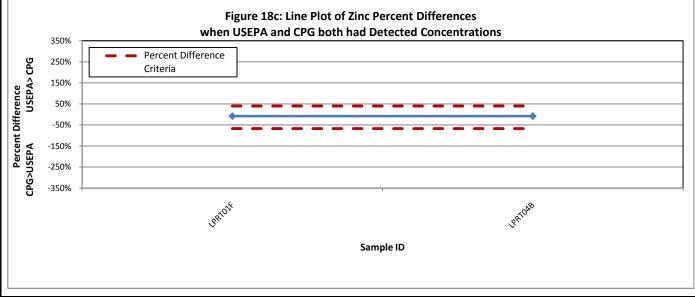




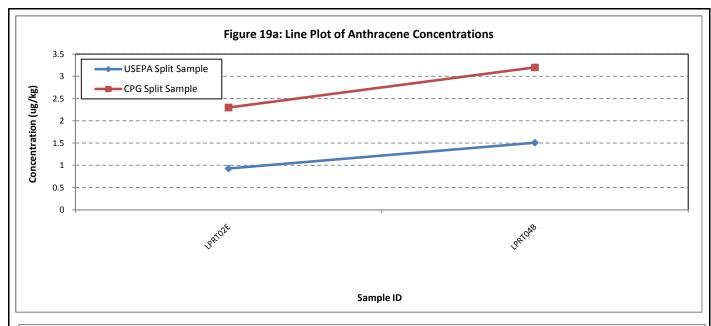


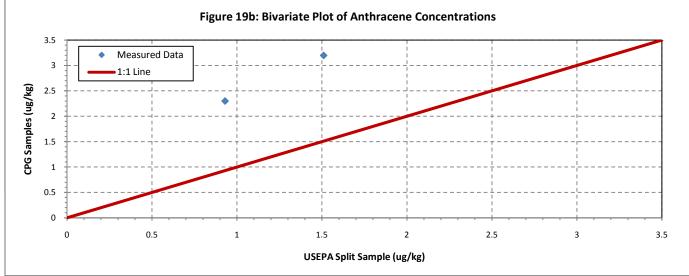


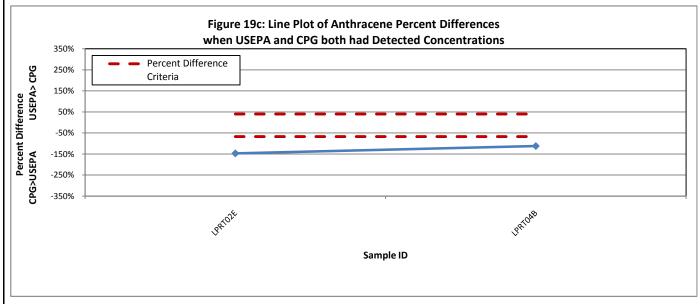




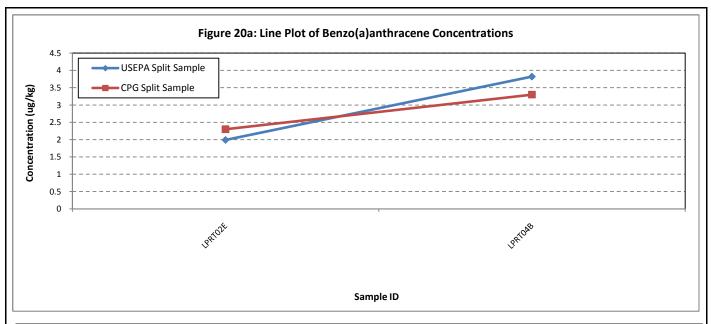


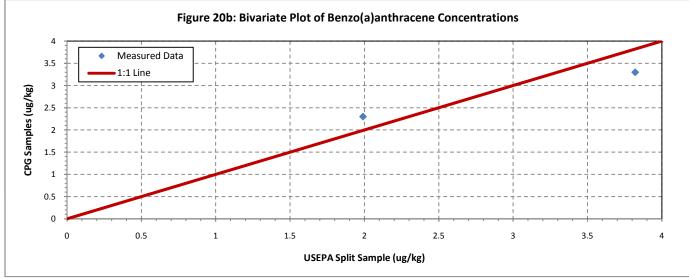


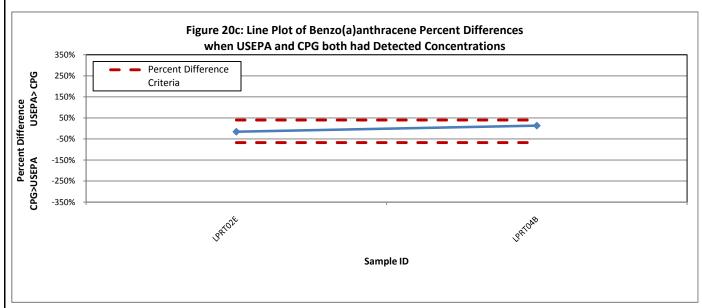




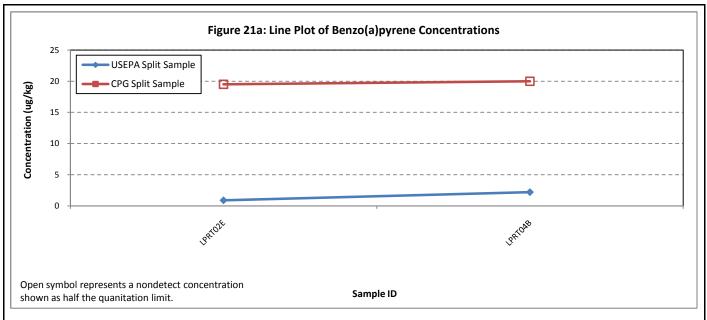


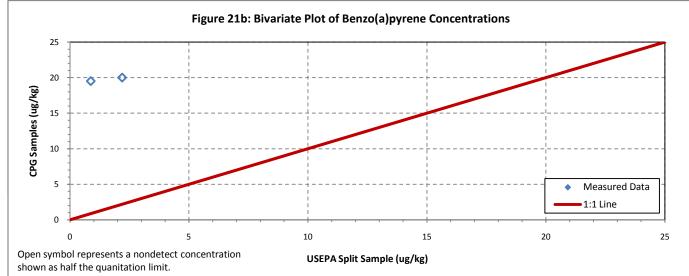


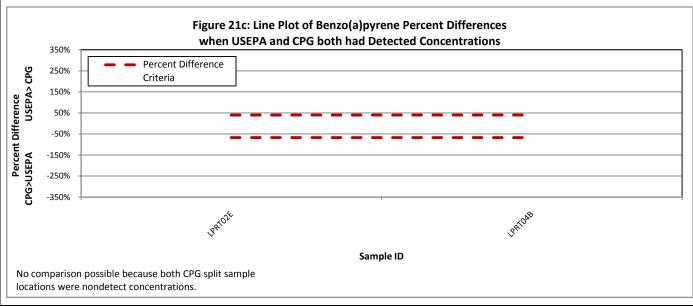






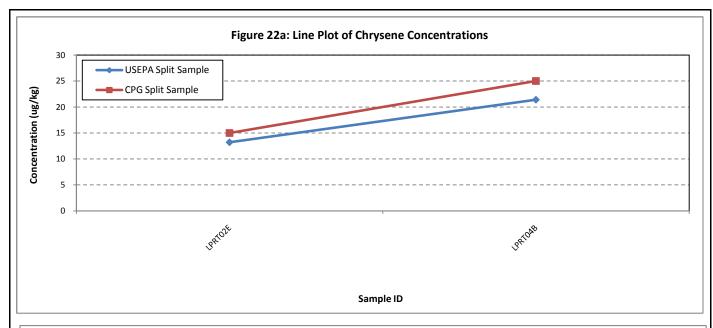


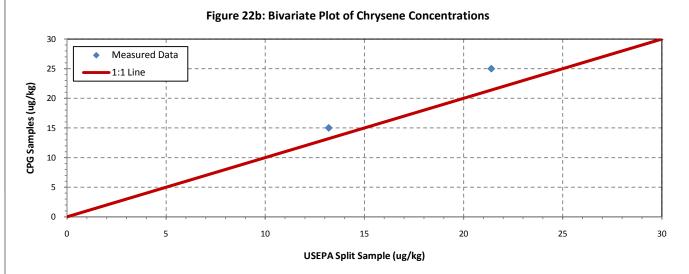


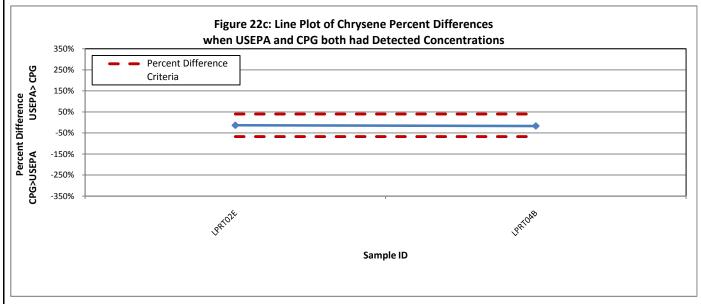




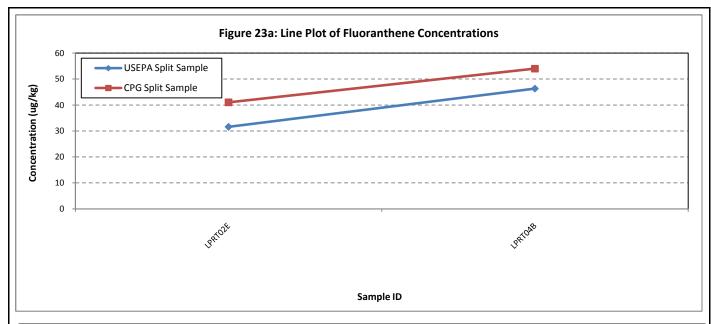


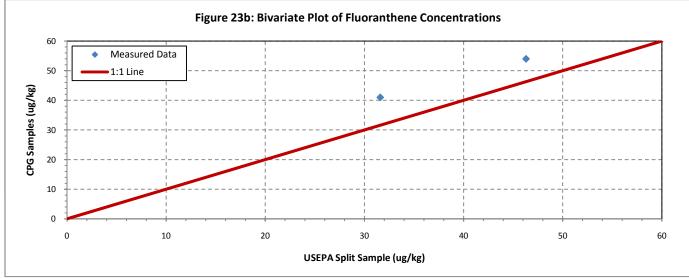


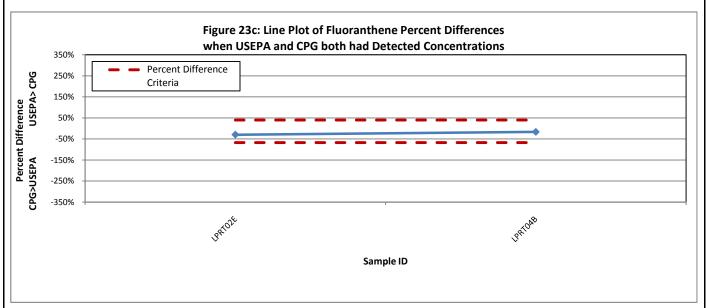




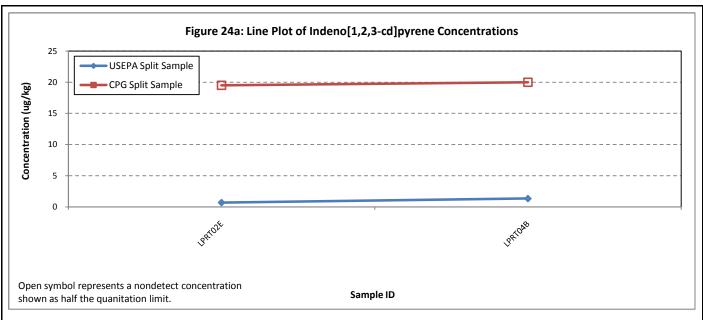


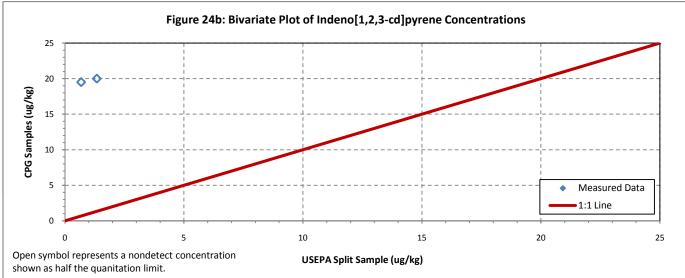


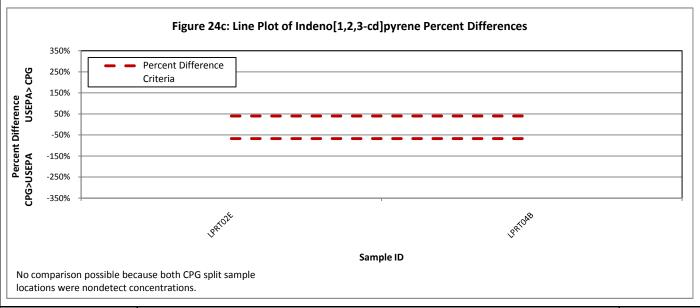




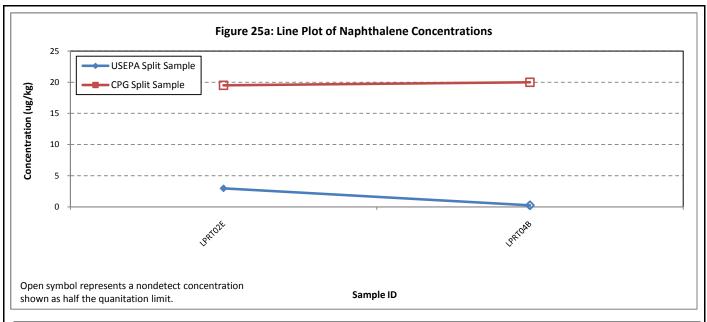


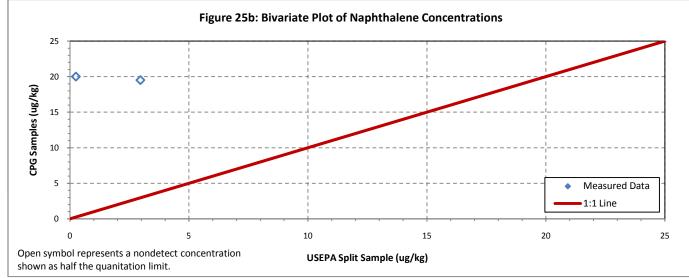


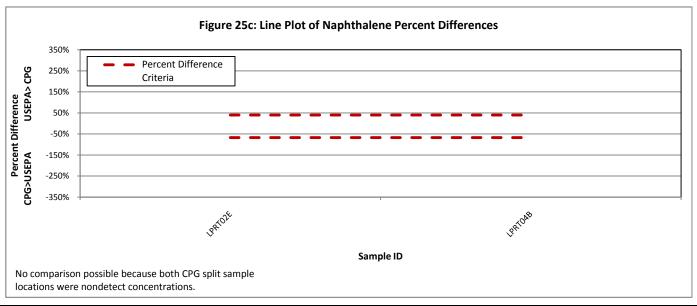




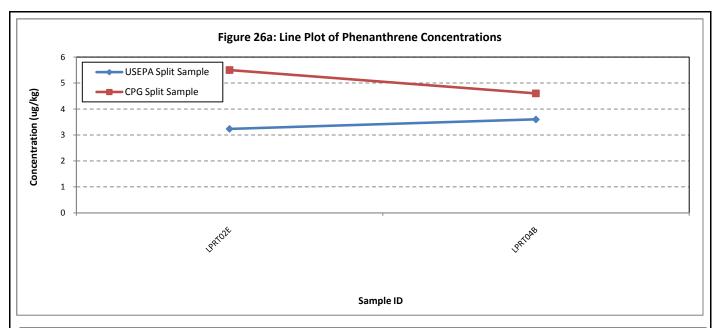


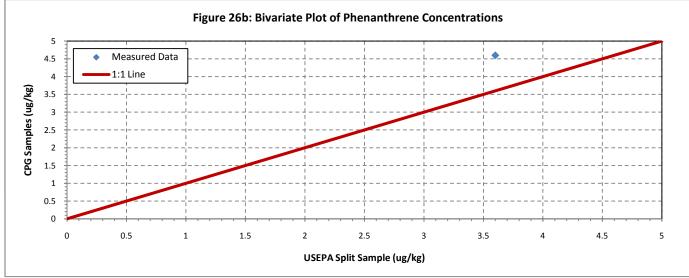


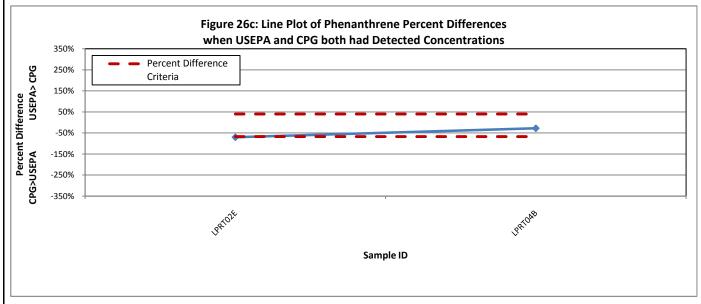




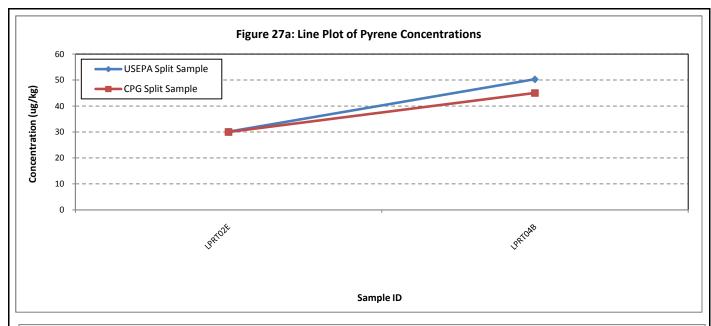


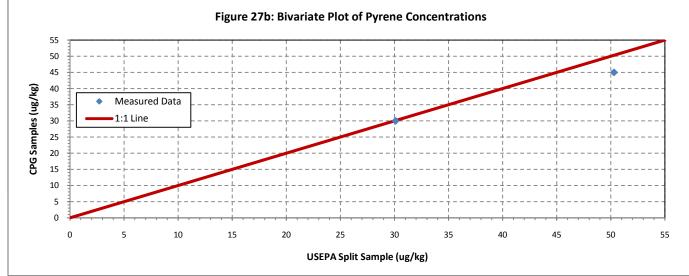


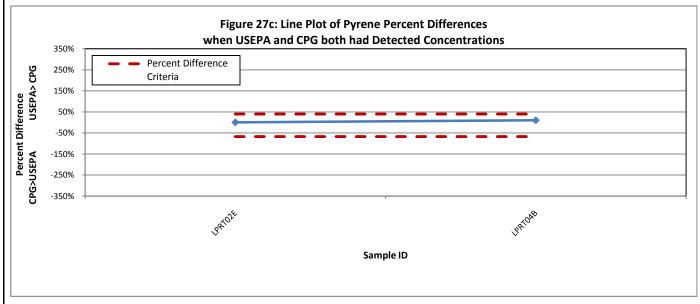




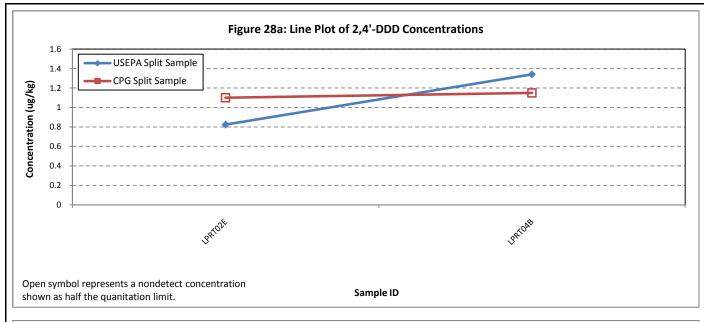


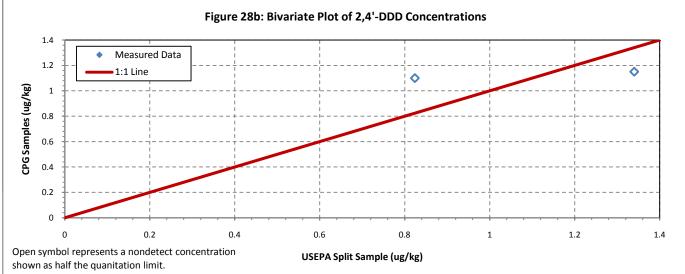


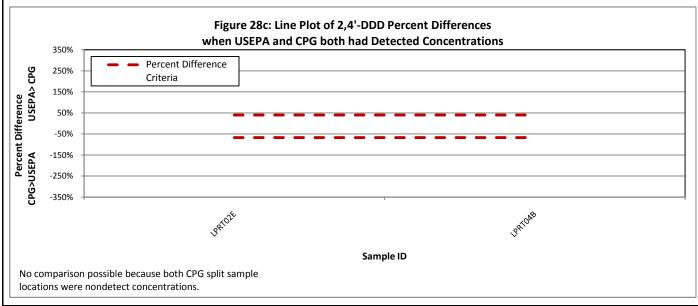




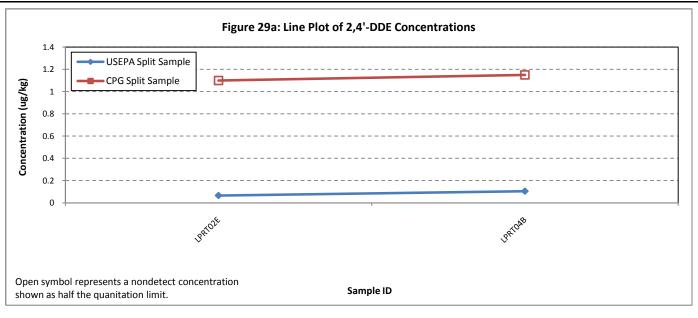


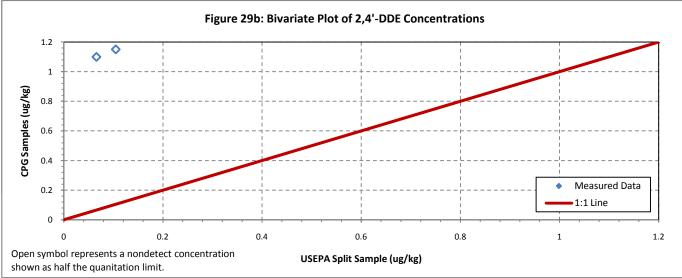


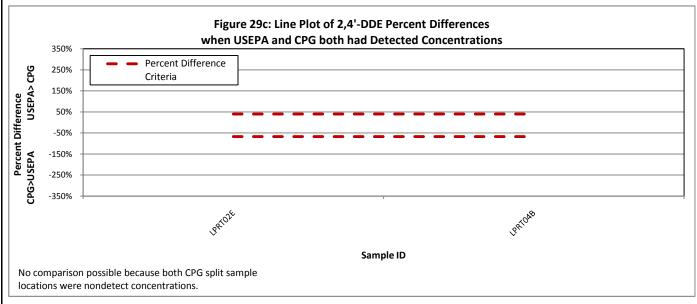




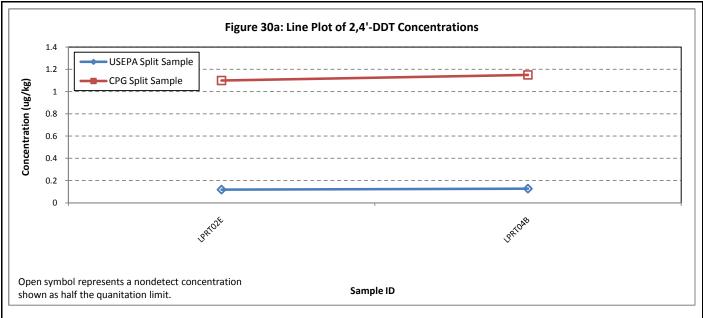


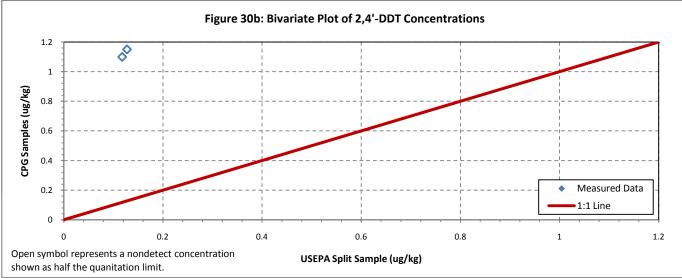


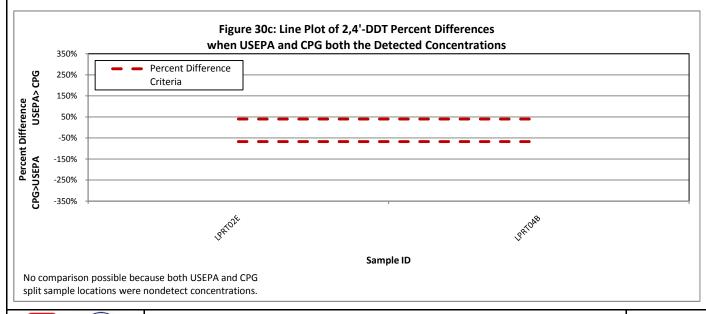




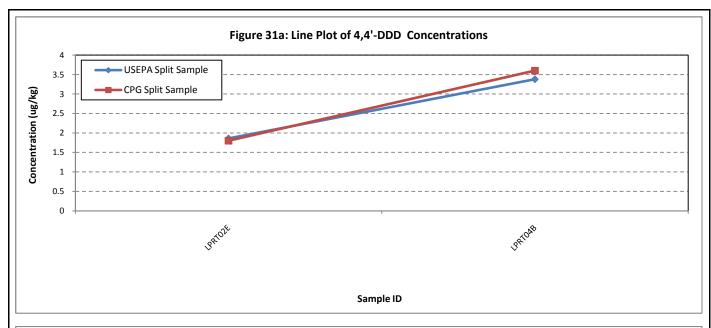


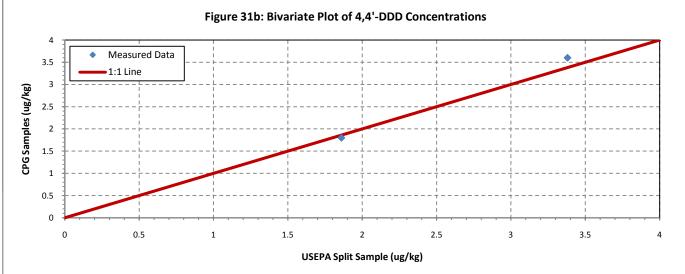


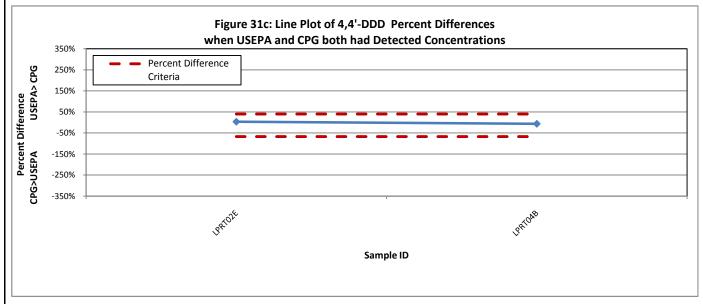


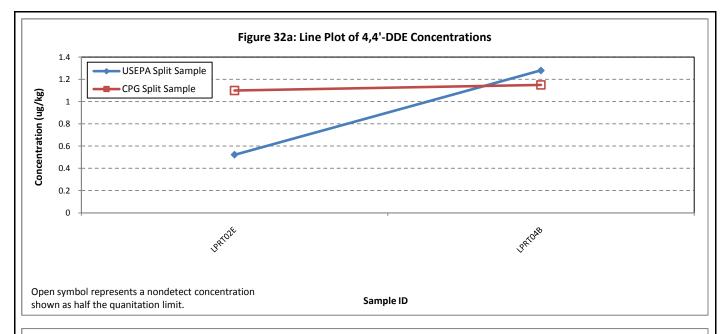


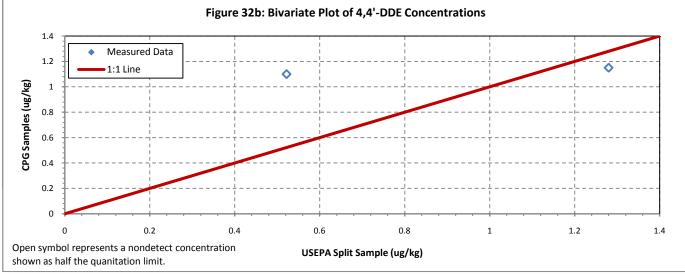


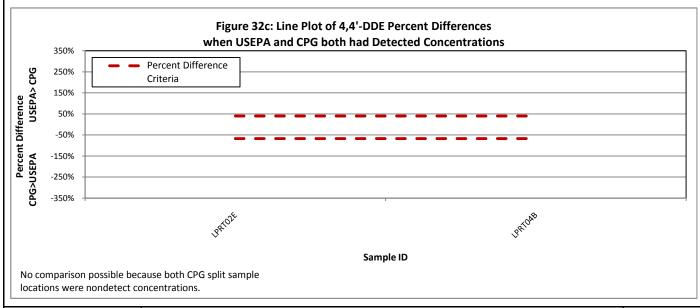




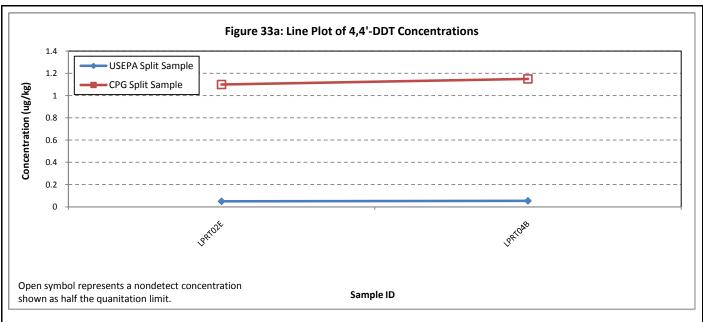


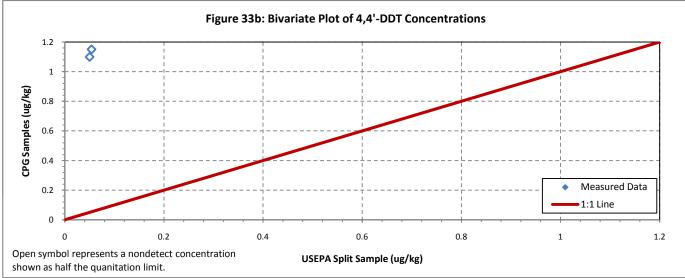


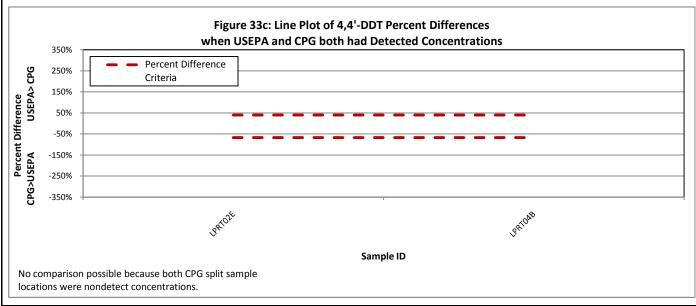




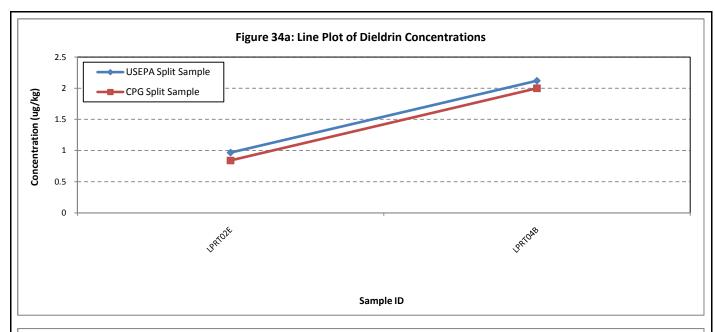


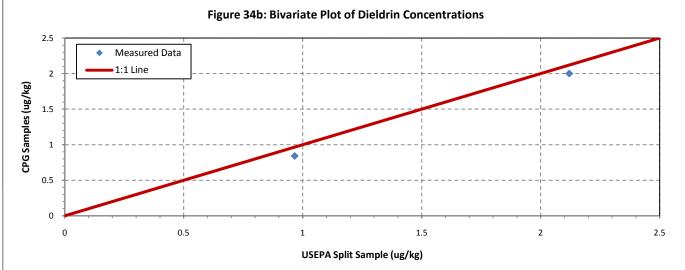


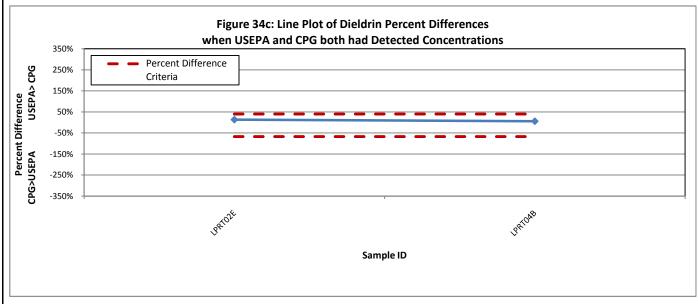




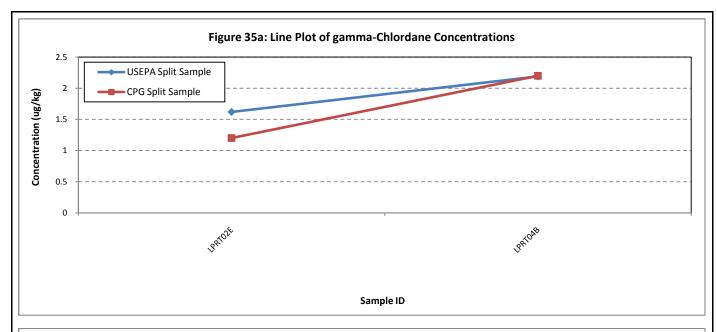


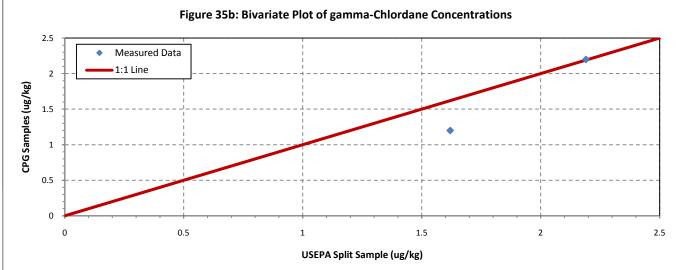


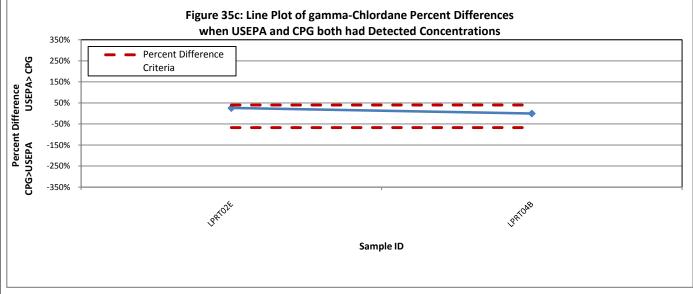




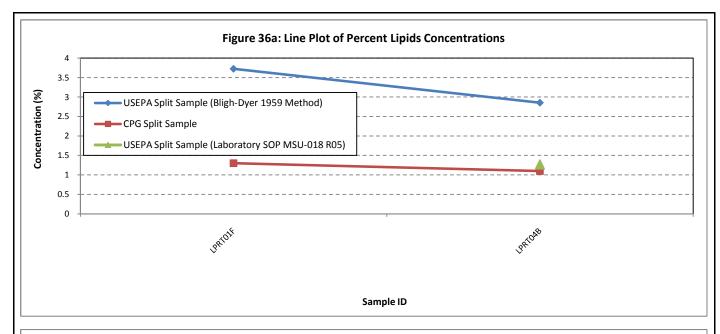


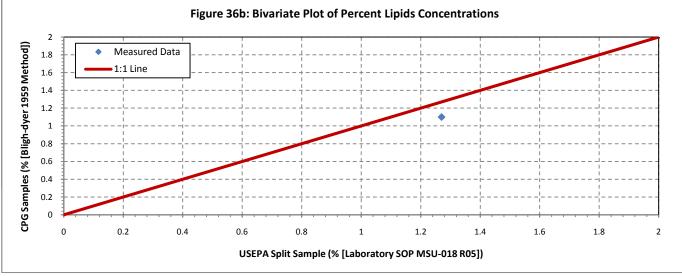


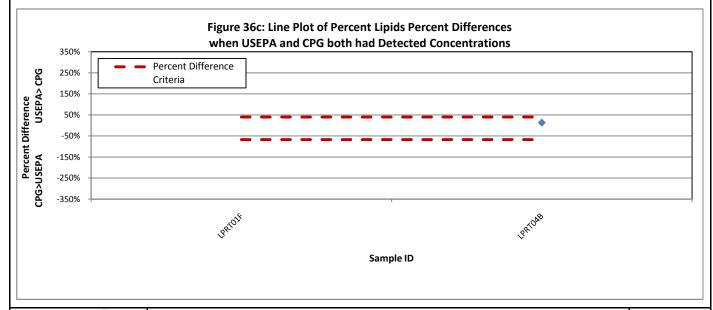


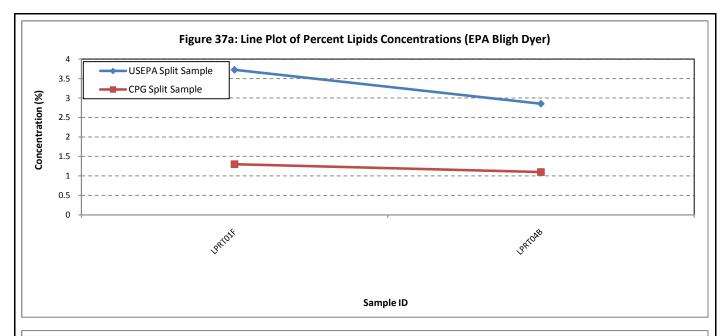


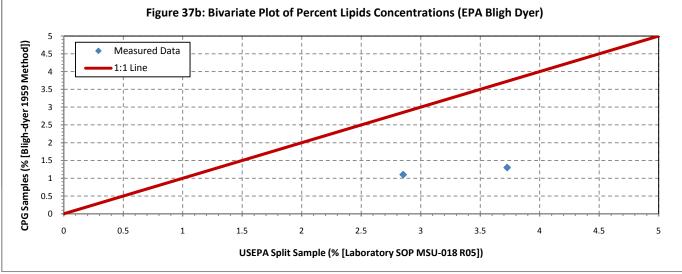


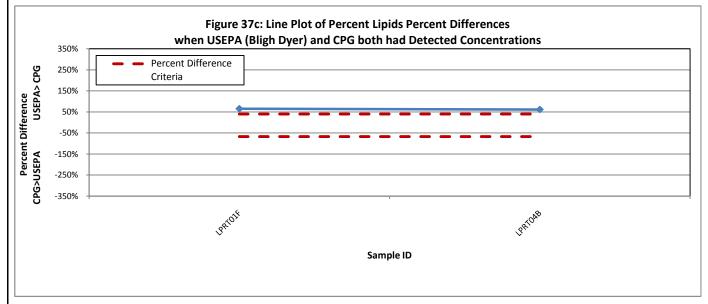




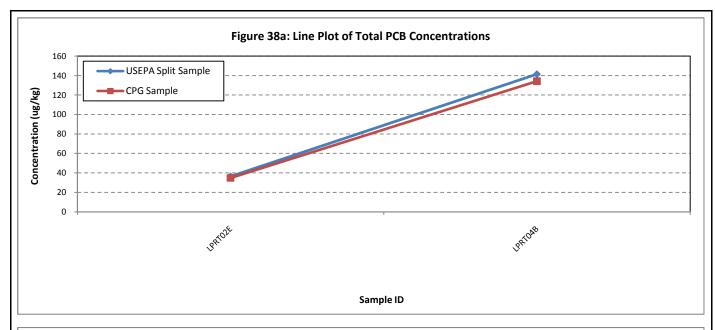


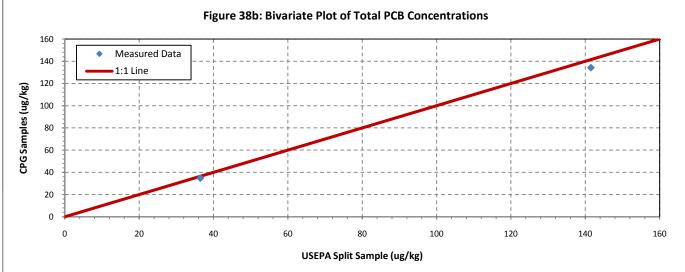


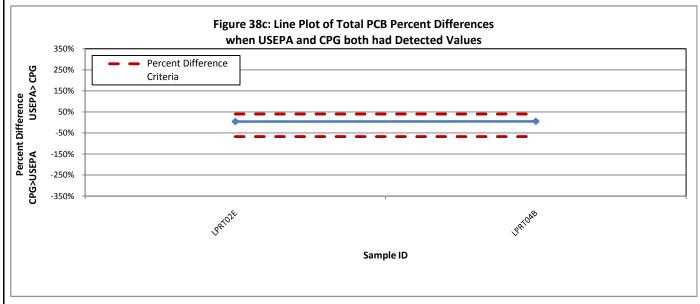




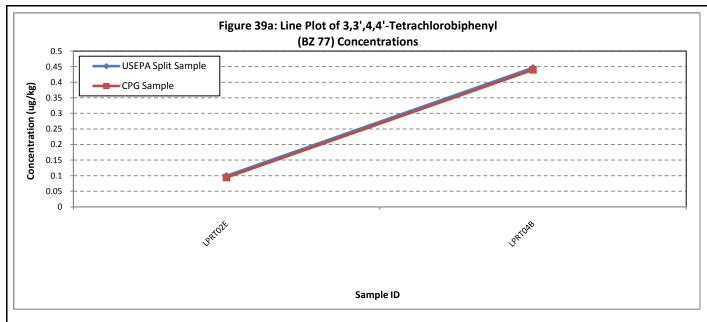


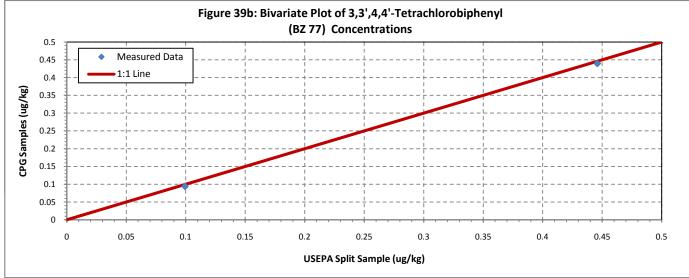


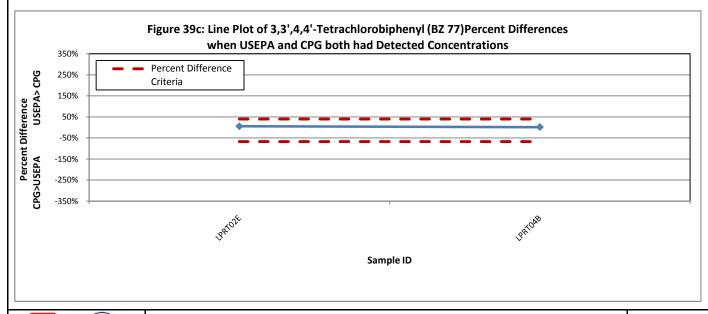




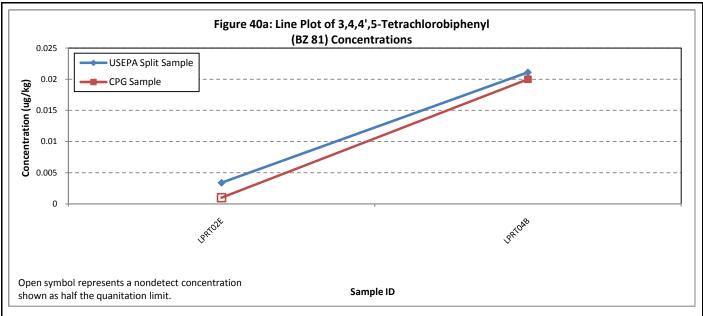


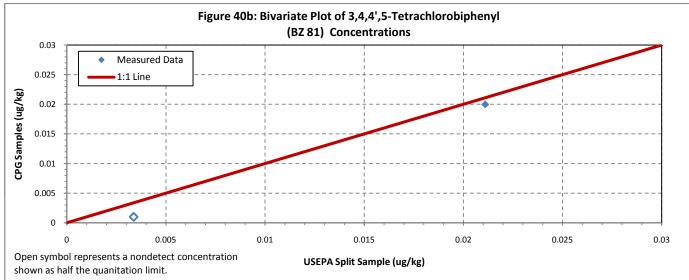


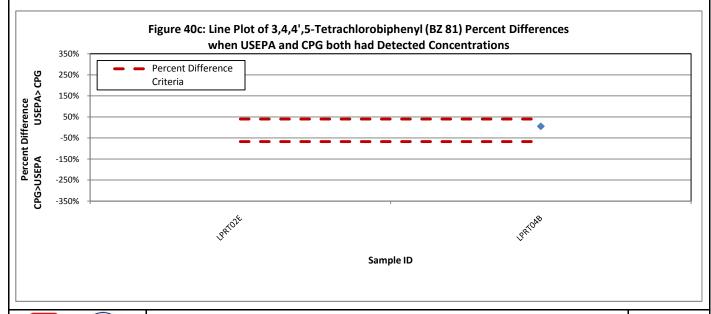




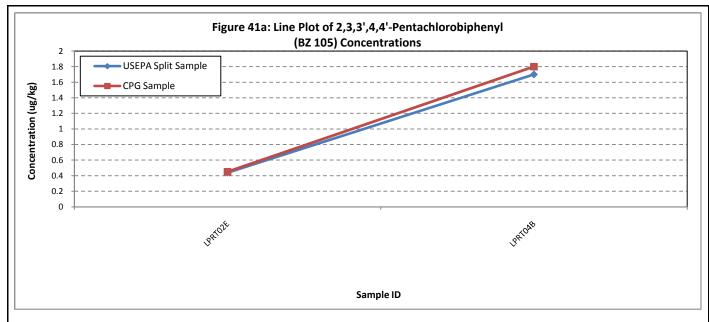


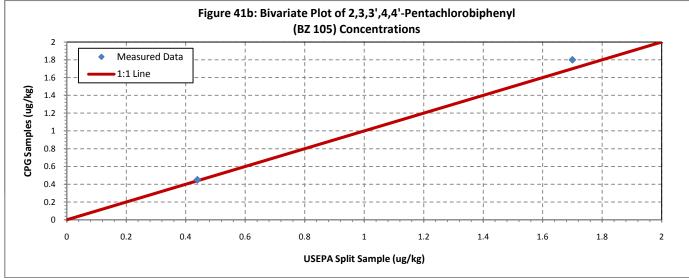


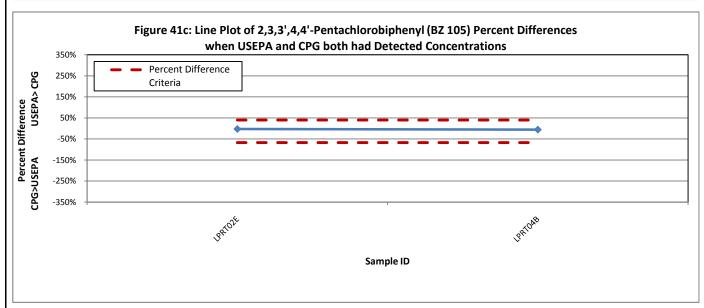




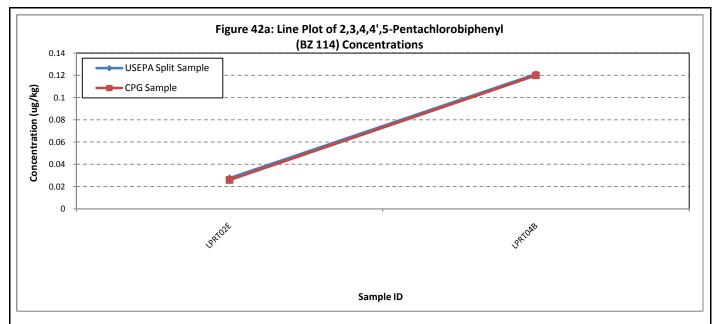


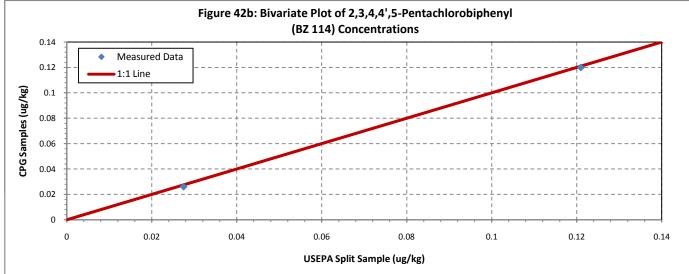


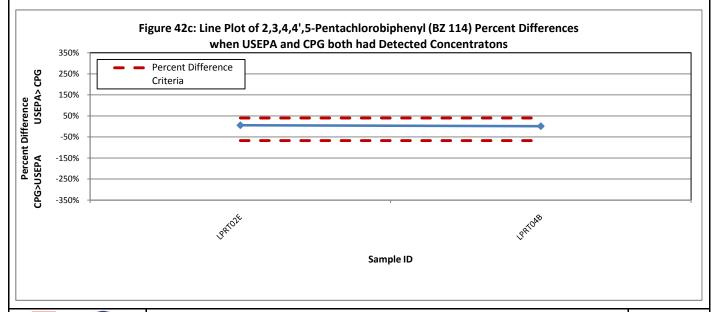




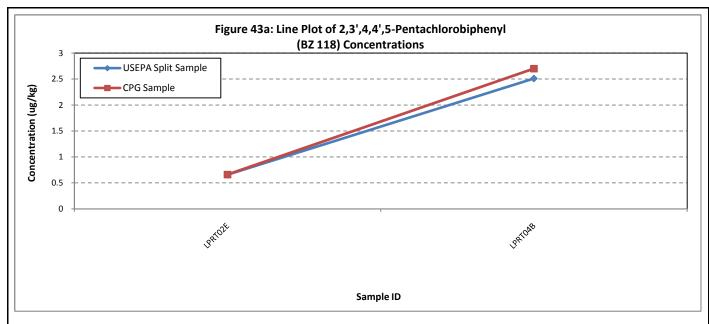


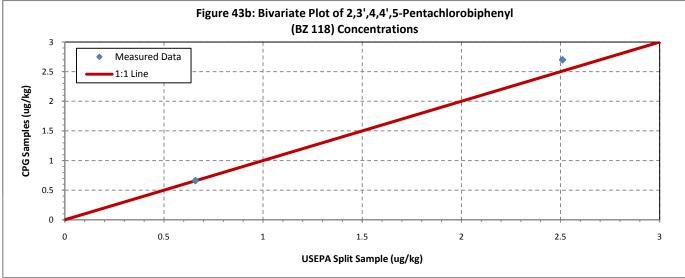


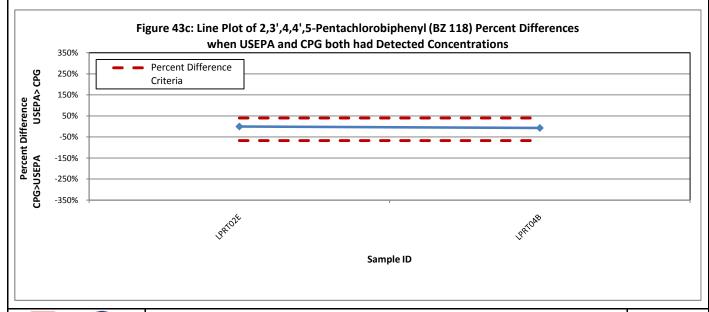




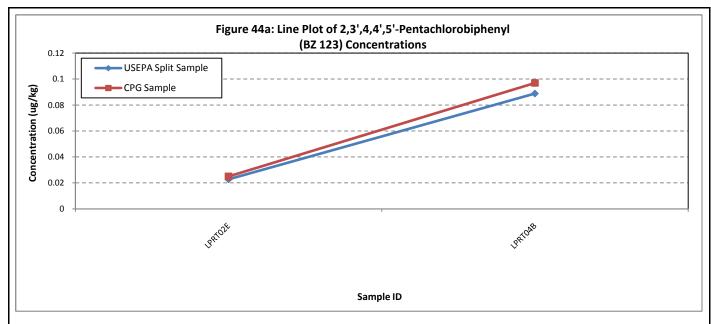


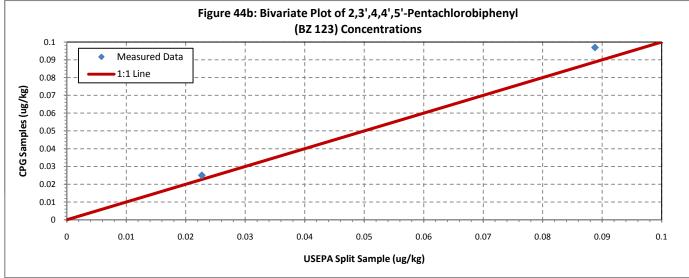


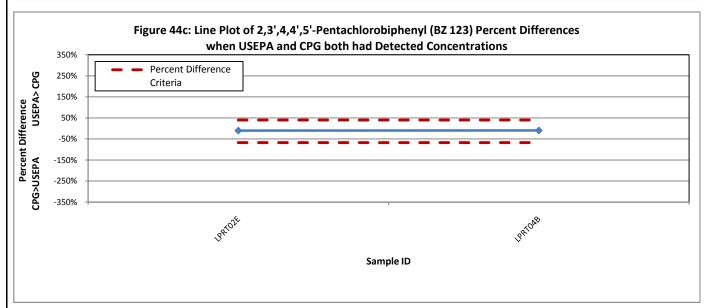




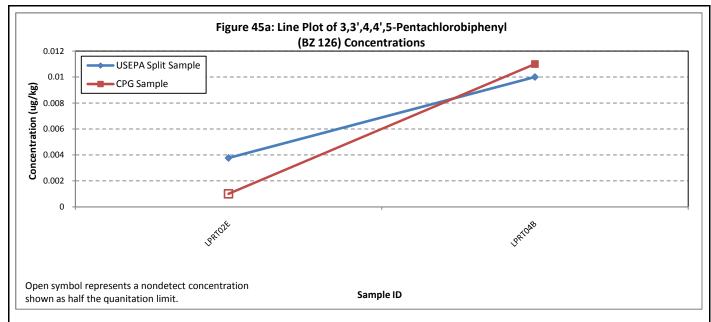


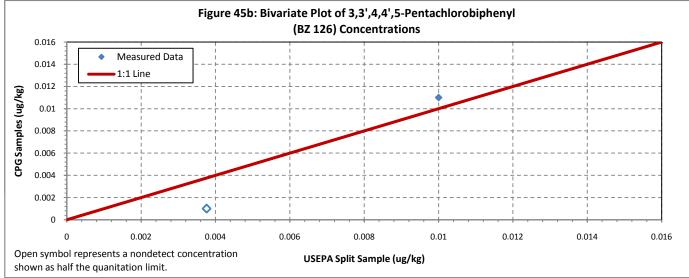


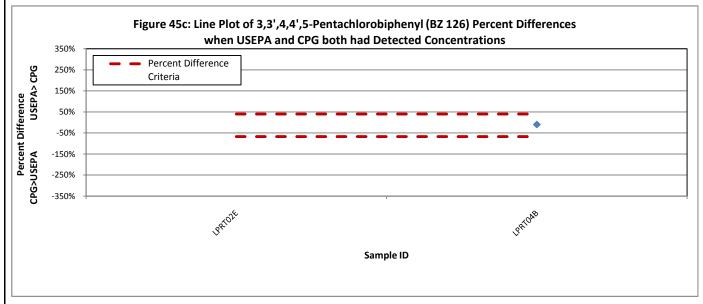




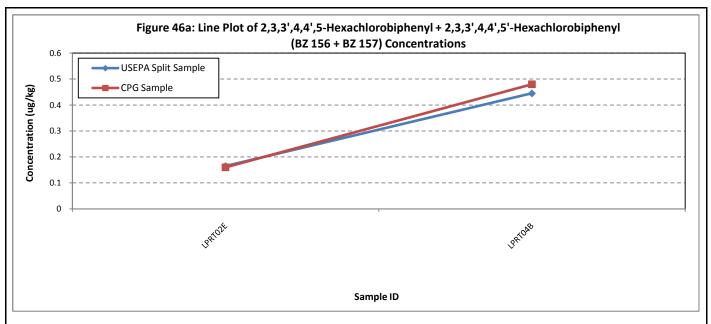


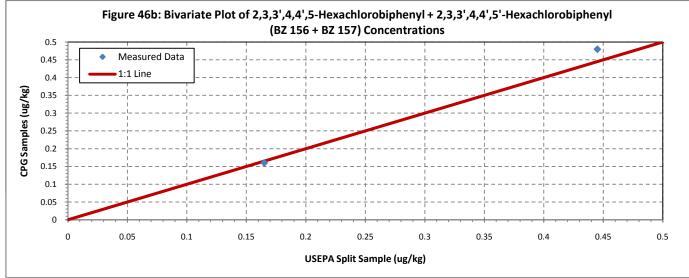


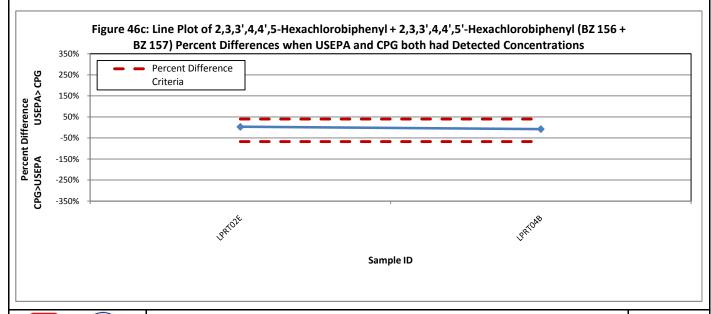




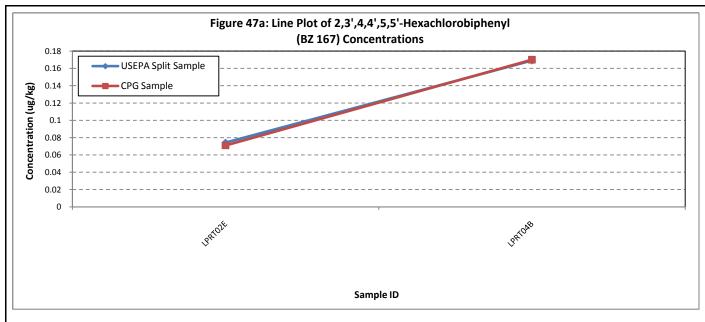


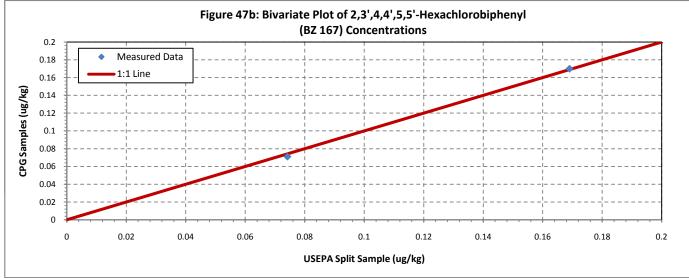


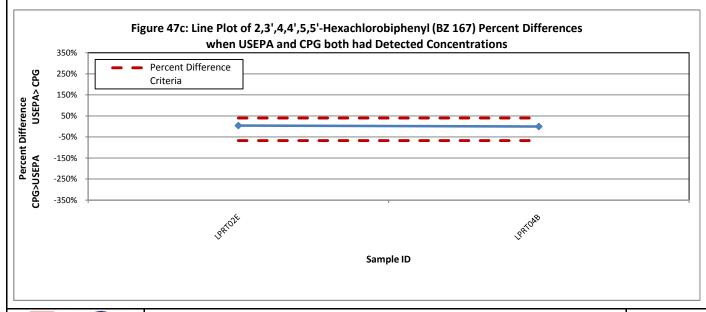




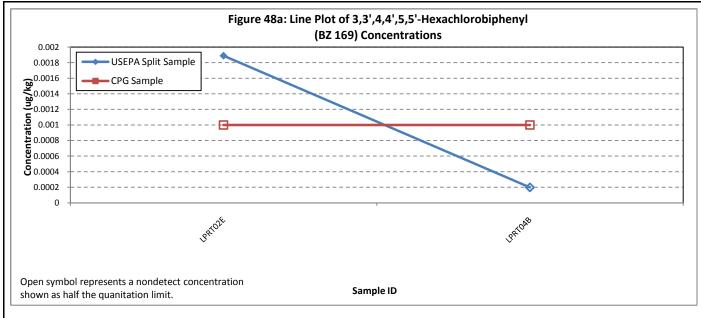


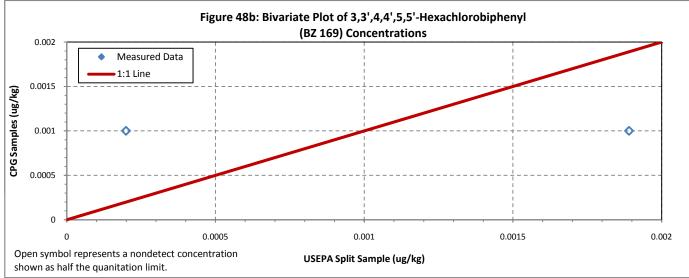


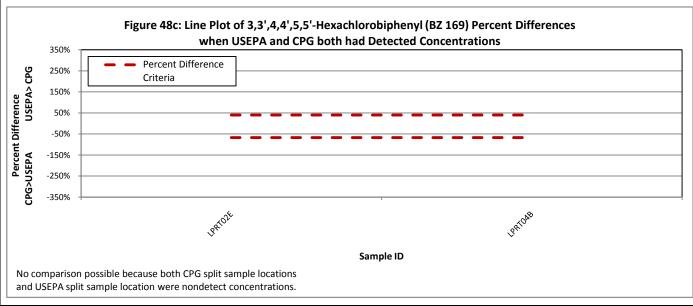




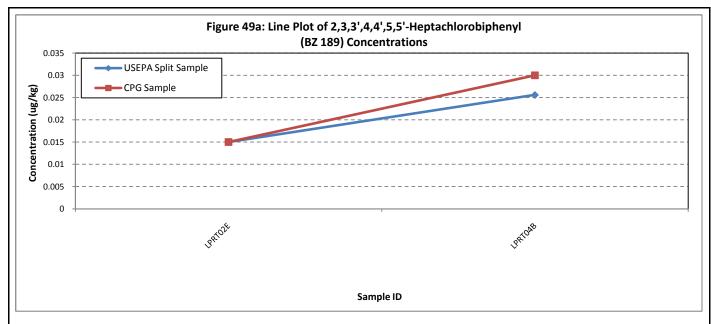


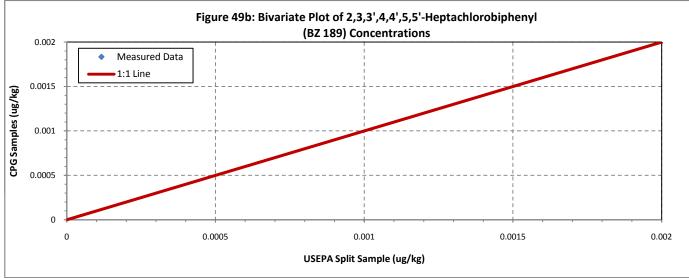


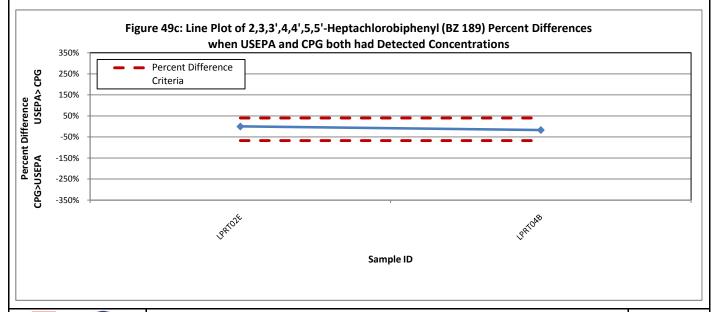














Battelle

Report

Toxicity Test Verification for Lower Passaic River Project



July 8, 2010

Dr. AmyMarie Accardi-Dey The Louis Berger Group, Inc. 565 Taxter Road, Suite 510 Elmsford, NY 10523

Subject: Toxicity Test Verification for Lower Passaic River Restoration Project

Dear AmyMarie:

Attached are verification reports for the review of four split sample sediment toxicity tests that were conducted by American Aquatic Testing, Inc. for the Lower Passaic River Restoration Project. The reports are formatted into three sections - Introduction, Verification Procedures, Verification Results, and Assessment of Usability, respectively. The detailed checklist used to guide the toxicity test verification is provided as Attachment 1 for each report. If you have any questions regarding this deliverable please contact Rosanna Buhl at 781-952-5309 or me at 631-941-3213.

Sincerely,

Elisabeth S. Barrows

Project/Program Manager

Jeky Barron

Attachments

cc: L. Warner (Berger); R. Buhl (Battelle); Battelle Records Management Office

Ampelisca abdita Toxicity Test for the Lower Passaic River Restoration Project

1.0 INTRODUCTION

During October 2009, sediment samples were collected at locations along the Passaic River as part of a Remedial Investigation/Feasibility Study (RI/FS) pursuant to the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA) and the Superfund Amendment and Reauthorization Act, as agreed to by the U.S. Environmental Protection Agency (EPA) and a group of 73 companies, the Cooperative Parties Group (CPG), considered potentially responsible for contamination in the lower Passaic River. On behalf of the U.S. Army Corps of Engineers (USACE) and U.S. EPA, Malcolm Pirnie, Inc. and its subcontractor, The Louis Berger Group, Inc., provided oversight and collected and analyzed government split samples. Government split sample data will be compared to the parent samples collected by the CPG to determine if a bias exists in the data produced by the CPG.

Sediment split sample toxicity testing was performed by American Aquatic Testing, Inc. (AAT) according to the *Oversight Quality Assurance Project Plan (QAPP) for Biological Sampling, Community Surveys, and Toxicity and Bioaccumulation Testing* dated August 6, 2009 and Field Modifications No. 2 (October 15, 2009) and No. 3 (December 23, 2009). Five *Ampelisca abdita* toxicity tests, representing amphipod exposure to estuarine sediments, were conducted by AAT for the Lower Passaic River Restoration Project.

2.0 VERIFICATION PROCEDURES

An independent verification of the *Ampelisca* toxicity test conditions and results was conducted by Battelle to verify that the test was conducted according to the QAPP and that the test results were acceptable. Acceptability of the toxicity test was assessed by comparing the AAT test procedures and conditions vs. the project requirements. Test procedures and results were described in the AAT report *Lower Passaic River Estuarine Section Restoration Project Sediment Toxicity Testing - Ampelisca abdita* (undated). The project requirements for the toxicity tests were defined in the following project control documents:

- Oversight Quality Assurance Project Plan (QAPP) for Biological Sampling, Community Surveys, and Toxicity and Bioaccumulation Testing (August 6, 2009)
- *QAPP* Field Modifications No. 2 (October 15, 2009)
- *QAPP* Field Modification No. 3 (December 23, 2009)
- Acute Toxicity of Sediments to the Marine Amphipod, *Ampelisca abdita* Project Specific Document (EnviroSystems, Inc. SOP QA-1426 Rev. 8c)

Toxicity test verification was initiated by identifying the test requirements defined in the above documents. In particular, the QAPP Worksheets (WSs) #36 and 37 define the acceptance criteria as those contained in WSs #12 and #28. In addition, The Louis Berger Group, Inc. statement of work indicated that tests should be verified vs. the QAPP, field modifications, revised toxicity SOPs, and issues encountered. The test requirements were tabulated in a checklist (Attachment 1), which was used to guide the review.

Ampelisca abdita Toxicity Test for the Lower Passaic River Restoration Project

3.0 VERIFICATION RESULTS

According to the QAPP, toxicity test acceptability is based on the health of the organisms and the acceptability of test conditions (WSs 12 and 28). The verification of these criteria is summarized below. The checklist provided as Attachment 1 details the full test verification results.

1. Health of Organisms (Laboratory negative control)

The health of organisms based on the laboratory negative control is verified as **acceptable**. Average negative control survival was 93% vs. the QAPP requirement of \geq 90%. Individual replicate survival ranged from 85 – 100% vs. the QAPP requirement of \geq 80%.

2. Health of Organisms (Laboratory positive control):

The health of organisms based on the laboratory positive control **cannot be determined**. A 48-hour KCl reference toxicant test was conducted but the results cannot be used to verify the health of the organisms because the laboratory does not typically run this positive control and therefore does not have historical control limits.

3. Acceptability of test conditions

The test conditions during the test are verified as **acceptable**. Water quality conditions met the criteria defined in the QAPP with minor exceptions.

- The dissolved oxygen (DO) concentration was maintained at ≥ 6.0 mg/L throughout the test with the following exceptions: the DO in three surrogate containers ranged between 5.5 and 5.9 mg/L on Day 0 prior to addition of the test organisms and fell to 5.8 mg/L on Day 2 in Sample LPRT02A. The QAPP states that dissolved oxygen concentrations must be ≥ 6.0 mg/L throughout the test. The test DO concentrations are **acceptable** because these minor deviations will not impact the test.
- The temperatures of overlying water in the test treatments ranged from 19.1 20.9°C throughout the test and are **acceptable**. The QAPP states that daily mean temperature must be within 20°C ±1°C. This criteria was achieved.
- Test salinity was maintained at 30±2 ppt throughout the test with two minor excursions above 32.0 ppt (32.1 and 32.3 ppt). The QAPP states that salinity concentrations must be 30±2 ppt throughout the test. These minor excursions from the defined salinity range do not impact the test; salinity conditions are **acceptable**.
- Water quality conditions during the test were monitored at the frequency specified in the QAPP and SOP. Water quality monitoring is judged to be **acceptable.**

4.0 ASSESSMENT OF USABILITY

The *Ampelisca abdita* test results are verified as **acceptable without reservation**. Holding times, negative control treatment survival, and water quality conditions met the QAPP criteria. The positive control results could not be used to assess animal health because the laboratory did not have historical data for comparison. Attachment 1 provides a full assessment of the toxicity test procedures and results vs. the QAPP requirements.

Data Quality Element	References	Verification Assessment
Test Design 1. Test approximately five sediments that are estuarine (≥5 ppt salinity)using the Ampelisca abdita 10-day survival toxicity test 2. Testing will follow EnviroSystems SOP QA-1426 Rev. 8c 3. A. abdita organisms for testing will be supplied by ARO; the same supplier used by EnviroSystems. 4. Artificial substrate for controls will be supplied by ARO and used to conduct one control sample test. 5. Seawater for controls will be supplied by ARO. 6. Sediment samples will not be sieved prior to testing.	WS#10 WS#11 WS#18 WS#19 WS#23 MOD#3¹	1. Yes, as modified by Field Modification #3. It was not possible to verify that the sediment samples tested using <i>Ampelisca abdita</i> were collected from an estuarine location because no data for the initial porewater salinity was provided in the report package. 2. Yes, as modified by Field Modification #3. Note: an additional, initial overlying water replacement that was not described in the SOP was conducted. 24 hours after sediment and overlying water was added to the test chambers, the overlying water was removed and new salt water was added to the sediment. The additional water replacement does not impact the test results because overlying water is renewed twice daily throughout the test. 3. Yes. The report narrative states that test organisms were supplied by ARO and were held under test conditions prior to testing. 4. No. The report narrative states that the control sample was tested using natural sediment provided by ARO. 5. Cannot be determined. The report narrative states that overlying water was prepared using natural saltwater (26 ppt) that was adjusted with dry sea salt to 30 ppt. The salt was provided by ESI. The narrative does not state that water was supplied by ARO. 6. No. The report narrative states that the samples were not sieved prior to testing. However, the raw data sheets document that the control sediment was sieved prior to testing. It is not acceptable for control treatments to be treated differently than
7. The results of the toxicity test will	WS#11	test treatments. 7. Yes. Significance vs. the control test was
be statistically compared to		determined using ANOVA and Dunnett's

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¹ Field Modification #3 specifies that the changes to Worksheet #23 defined in the modification are applicable to split sample toxicity testing conducted after November 11, 2009. *Ampelisca abdita* toxicity testing was initiated on November 5, 2009, however, several of the modifications were discussed during a meeting conducted on October 21, 2009.

Data Quality Element	References	Verification Assessment
comparable tests conducted with control sediment for control survival.		pairwise comparisons.
8. Toxicity tests will be conducted according to the government assigned lab SOPs, modified so that test conditions are comparable to the CPG assigned laboratory SOP.	WS#11 MOD#3	8. Yes, as modified by Field Modification #3,except as noted elsewhere in this checklist.
Health of Test Organisms via laboratory negative control: 9. Average survival: ≥ 90% 10. Individual replicate survival: ≥ 80%	WS#12 WS# 28	 9. Yes. Average survival was 93%. 10. Yes. Individual replicate survival ranged from 85 – 100%.
Health of Test Organisms via laboratory positive control (reference toxicant): 11. A standard reference toxicity test will be conducted. 12. The LC50 for a positive control test should be within the mean LC50 ±2 standard deviations of the control chart.	WS#12 WS# 28 MOD#3	 11. Yes. A 48-hour KCl reference toxicant test was conducted. 12. Cannot be determined. The LC50 for the 48-hour KCl reference toxicant test was 1067.7 ppm. The health of test organisms could not be determined because the laboratory does not typically run this positive control and therefore does not have historical control limits.
Acceptability of test conditions: 13. Dissolved oxygen: ≥ 6.0 mg/L 14. Temperature (daily mean): 20°C² ±1°C 15. Salinity: 30±2 ppt 16. Monitoring Requirements: Water Quality Parameter: Dissolved oxygen, temperature, pH, and salinity. Frequency: Monitor in every test vessel at test start and end; daily during test in surrogate test vessel for each treatment. Water Quality Parameter: Overlying and porewater ammonia. Frequency: Monitor in surrogate test vessel at test start, day 3, and end.	WS#12 WS# 28 MOD#3 SOP QA- 1426 Rev. 8c	 13. Yes. The dissolved oxygen (DO) concentration was maintained at ≥ 6.0 mg/L throughout the test with the following exceptions: the DO in three surrogate containers ranged between 5.5 and 5.9 mg/L on Day 0 prior to addition of the test organisms and fell to 5.8 mg/L on Day 2 in Sample LPRT02A. These minor deviations will not impact the test. 14. Yes. The temperatures of overlying water in the test treatments ranged from 19.1 – 20.9°C throughout the test. 15. Test salinity was maintained at 30±2 ppt throughout the test with two minor excursions above 32.0 ppt (32.1 and 32.3 ppt). These minor excursions from the defined salinity range do not impact the test. 16. Yes. Water quality conditions during

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 $^{^2}$ Note that the test temperature was changed from 15 to 20°±1°C in the SOP modified with Field Modification #3.

Data Quality Element	References	Verification Assessment
		the test were monitored at the frequency specified in the QAPP and SOP.
Test conditions: 17. Unionized ammonia <0.4 mg/L 18. Five replicates with 20 amphipods/replicate chamber 19. Immature amphipods, 3-5 mm; no reproductive adults	SOP QA- 1426 Rev. 8c	 17. Yes. The raw data states that total ammonia values were too low for calculation of unionized ammonia. 18. Yes. 19. Yes. The report narrative states that at the beginning of the test organisms were adolescents 3-5 mm long.
 Sample Handing 20. Preservation: ≤ 4 degrees Celsius 21. Holding Time: ≤8 weeks, preferably ≤14 Days 22. All toxicity testing will be performed using the same two gallons of unsieved sediment. 23. Samples will not be sieved prior to testing. 24. Project sediments will be stored at 2-4°C and will not be purged with inert gas once opened. 	WS#19 MOD#3	 20. Cannot be determined. According to the report narrative, sediments were collected on October 13 and 14, 2009 and received on ice at AAT on October 16, 2009. The temperature of the sediments upon receipt was not provided in the report. 21. Yes. Sample testing began on November 5, 2009, 23 days after sample collection. 22. Cannot be determined. The report narrative does not state that all toxicity testing was conducted using the same sediment samples (i.e., both <i>Ampelisca</i> and <i>Chironomus</i>). However, the custody forms identified that samples were to be used for testing both species. 23. No. The narrative confirms that test sediments were not sieved. However, according to the raw data sheets, the control sediment was sieved prior to use. 24. Yes. Upon receipt the samples were refrigerated until testing was initiated on November 5, 2009. Comment on sample traceability: Five sediment samples were tested (LPRT02F, LPRT03A, LPRT01F, LPRT02A, and LPRT01G). The report package did not include the custody forms for these samples. Accutest chain of custody

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Data Quality Element	References	Verification Assessment
		forms were included in the data package for three AQ samples (09839, 09841, and 09842) and two soil samples (09843 and 09844). Based on the report package, there is no mechanism to match the custody form sample identification numbers to the reported sample values. No custody forms were provided for the test organisms or sea salt.
Delivery 25. Data turn-around time: 90 days (60 for testing and 30 for validation)	WS#30	25. Not assessed . The data report is not dated.
Validation	WS#36	26. Yes. Completed as specified.
26. Toxicity testing data will not require full data validation. Toxicity data will only be reviewed against the acceptance limits provided in Worksheets 12 and 28.		
Usability	WS#37	27. Yes. Holding times, negative control
27. Usability of toxicity data is based on achieving sample holding times, acceptable water quality conditions during testing, and laboratory control treatment survival and growth criteria (sic - growth criteria are not applicable to the <i>Ampelisca</i> test).		treatment survival, and water quality conditions met QAPP criteria. The positive control results could not be used to assess animal health because the laboratory did not have historical data for comparison.

Chironomus dilutus Toxicity Test for the Lower Passaic River Restoration Project

1.0 INTRODUCTION

During October 2009, sediment samples were collected at locations along the Passaic River as part of a Remedial Investigation/Feasibility Study (RI/FS) pursuant to the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA) and the Superfund Amendment and Reauthorization Act, as agreed to by the U.S. Environmental Protection Agency (EPA) and a group of 73 companies, the Cooperative Parties Group (CPG), considered potentially responsible for contamination in the lower Passaic River. On behalf of the U.S. Army Corps of Engineers (USACE) and U.S. EPA, Malcolm Pirnie, Inc. and its subcontractor, The Louis Berger Group, Inc., provided oversight and collected and analyzed government split samples. Government split sample data will be compared to the parent samples collected by the CPG to determine if a bias exists in the data produced by the CPG.

Sediment split sample toxicity testing was performed by American Aquatic Testing, Inc. (AAT) according to the *Oversight Quality Assurance Project Plan (QAPP) for Biological Sampling, Community Surveys, and Toxicity and Bioaccumulation Testing* dated August 6, 2009 and Field Modifications No. 2 (October 15, 2009) and No. 3 (December 23, 2009). Five *Chironomus dilutus* toxicity tests, representing midge larvae exposure to freshwater sediments, were conducting by AAT for the Lower Passaic River Restoration Project.

2.0 VERIFICATION PROCEDURES

An independent verification of the *Chironomus* toxicity test conditions and results was conducted by Battelle to verify that the test was conducted according to the QAPP and that the test results were acceptable. Acceptability of the toxicity test was assessed by comparing the AAT test procedures and conditions vs. the project requirements. Test procedures and results were described in the AAT report *Lower Passaic River Freshwater Section Restoration Project Sediment Toxicity Testing - Chironomus dilutus* (undated). The project requirements for the toxicity tests were defined in the following project control documents:

- Oversight Quality Assurance Project Plan (QAPP) for Biological Sampling, Community Surveys, and Toxicity and Bioaccumulation Testing (August 6, 2009)
- *OAPP* Field Modifications No. 2 (October 15, 2009)
- *QAPP* Field Modification No. 3 (December 23, 2009)
- Acute Toxicity of Sediments to the Midge Larvae, *Chironomus dilutus* Project Specific Document (EnviroSystems, Inc. SOP QA-1407 Rev. 12c)

Toxicity test verification was initiated by identifying the test requirements defined in the above documents. In particular, the QAPP Worksheets (WS) #36 and 37 define the acceptance criteria as those contained in WSs #12 and #28. In addition, The Louis Berger Group, Inc. statement of work indicated that tests should be verified vs. the QAPP, field modifications, revised toxicity SOPs, and issues encountered. The test requirements were tabulated in a checklist (Attachment 1), which was used to guide the review.

Chironomus dilutus Toxicity Test for the Lower Passaic River Restoration Project

3.0 VERIFICATION RESULTS

According to the QAPP, toxicity test acceptability is based on the health of the organisms and the acceptability of test conditions (WSs 12 and 28). The verification of these criteria is summarized below. The checklist provided as Attachment 1 details the full test verification results.

1. Health of Organisms (Laboratory negative control)

The health of organisms based on the laboratory negative control is verified as **acceptable**. Average negative control survival was 93.8% vs. the QAPP requirement of $\geq 70\%$.

The health of organisms based on average ash free dry weight of surviving organisms is determined to be **unacceptable.** The control treatment average ash free dry weight was 0.425 mg vs. the QAPP requirement of ≥ 0.48 mg per surviving individual. It is noted that all test treatment growth rates exceeded the average ash free dry weight requirements, ranging from 0.516-0.731 mg.

2. Health of Organisms (Laboratory positive control):

The health of organisms based on the laboratory positive control **is acceptable**. The 48-hour KCl toxicant test LC50 (6830.2 ppm) was within the laboratory historical control chart limits.

3. Acceptability of test conditions

The test conditions during the test are verified as **acceptable**, with the exception of hardness which is **Possibly Not Acceptable**.

- Dissolved oxygen concentrations were >3.3 mg/L throughout the test and are **acceptable**. The QAPP states that dissolved oxygen concentrations must be ≥ 2.5 mg/L throughout the test.
- Temperatures of overlying water ranged from 21.6 − 24.0°C throughout the test and are **acceptable**. The QAPP states that daily mean temperature must be within 23°C ±1°C, no temperature value may exceed 23°C ±3 °C of the mean at any time, and the instantaneous temperature must always 23°C±3°C. All QAPP criteria were achieved.
- Alkalinity concentration differences between test initiation and termination ranged between 14 and 50% and are **acceptable**. The QAPP states that alkalinity concentrations should not vary by more than 50% during the test. All QAPP criteria were achieved.
- Hardness concentration differences between test initiation and termination ranged between 25 and 68%. The QAPP states that hardness concentrations should not vary by more than 50% during the test. In two treatments (LPRT11A and LPRT11D) hardness dropped by more than 50% (68 mg/L and 57 mg/L, respectively). The hardness conditions for these two samples are unacceptable. As discussed in the checklist (Attachment 1), this drop in hardness is unusual and should be further examined by the testing laboratory. Changes in hardness will impact the bioavailability of metals to the organisms.

Chironomus dilutus Toxicity Test for the Lower Passaic River Restoration Project

- Ammonia concentration differences between test initiation and termination ranged between 58 and 100% throughout the test and are **acceptable** despite exceedences from QAPP criteria. The QAPP states that ammonia concentrations should not vary by more than 50% during the test. However, because the ammonia concentrations are very low and not harmful at the measured levels (0 − 2.1 mg/L), these decreases are likely artifacts of the sediment characteristics and will not impact test acceptability.
- Water quality conditions during the test were monitored at the frequency specified in the QAPP and SOP with the exception that it was not possible to determine from the raw data if porewater ammonia and pH were measured in each test chamber at the end of the test. Water quality monitoring is judged to be acceptable.

4.0 ASSESSMENT OF USABILITY

The *Chironomus dilutus* test results are verified as **acceptable with reservations.** Holding times, positive and negative control treatment survival, and all water quality criteria except hardness in two samples met QAPP criteria. The ash free dry weight for the negative control and the degree of change in hardness between test initiation and termination in two samples were not acceptable. Attachment 1 provides a full assessment of the toxicity test procedures and results vs. the QAPP requirements.

Attachment 1 Lower Passaic River Restoration Project Chironomus dilutus 10-Day Survival and Growth Toxicity Test SOP QA-1407 Rev. 12c

Data Quality Element	References	Verification Assessment	
Test Design 1. Test approximately five sediments that are freshwater (<5 ppt salinity); using the <i>Chironomus dilutus</i> 10-day survival and growth toxicity test	WS#10 WS#11 WS#18 WS#19 WS#23 MOD#3 ²	1. Yes, as modified by Field Modification #3. It was not possible to verify that the sediment samples tested using <i>Chironomus dilutus</i> were collected from a freshwater location because no data for the initial porewater salinity was provided	
2. Testing will follow EnviroSystems SOP QA-1407 Rev. 12c	WIOD#3	in the report package.	
3. <i>C. dilutus</i> organisms for testing will be purchased from the same supplier used by EnviroSystems (either ABS, Inc. Fort Collins, CO. or ARO, Inc. Hampton NH). ¹		2.	2. Yes, as modified by Field Modification #3. Note: an additional, initial overlying water replacement that was not described in the SOP was conducted. 24 hours after sediment and overlying water was added to the test chambers, the overlying water
4. Artificial substrate for controls will be supplied by ARO and used to conduct one control sample test.		was removed and new fresh water was added to the sediment. The additional water replacement does not impact the	
5. EnviroSystems, Inc. will provide freshwater to AAT.		test results because overlying water is renewed twice daily throughout the test.	
6. Sediment samples will not be sieved prior to testing.		3. Yes. The report narrative states that test organisms were supplied by ABS and were held under test conditions prior to testing.	
		4. Yes. The report narrative states that the control sample was tested using artificial sediment provided by EnviroSystems.	
		5. Yes. The report narrative states that overlying water was natural freshwater provided by EnviroSystems. However, the report also states that overlying water was "created using natural fresh water provided by ESI and reconstituted fresh water prepared by AAT. These two statements appear to be contradictory.	
		6. Yes. The report narrative and raw data indicate that sediment was not sieved.	
7. The results of the toxicity test will be statistically compared to comparable tests conducted with control sediment for control survival and/or growth.	WS#11	7. Yes. Significance vs. the control test was determined using ANOVA and Dunnett's pairwise comparisons.	

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 $^{^{1}\} Field\ Modification\ No.\ 3\ lists\ the\ supplier\ as\ ABS\ under\ modifications\ to\ WS\#9\ and\ as\ ARO\ under\ {\it Chironomus}\ modifications\ to\ WS\#23.$

² Field Modification #3 specifies that the changes to Worksheet #23 defined in the modification are applicable to split sample toxicity testing conducted after November 11, 2009. *Ampelisca abdita* toxicity testing was initiated on November 5, 2009, however, several of the modifications were discussed during a meeting conducted on October 21, 2009.

Attachment 1

Lower Passaic River Restoration Project Chironomus dilutus 10-Day Survival and Growth Toxicity Test SOP QA-1407 Rev. 12c

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Data Quality Element	References	Verification Assessment
8. Toxicity tests will be conducted according to the government assigned lab SOPs, modified so that test conditions are comparable to the CPG assigned laboratory SOP.	WS#11 MOD#3	8. Yes, as modified by Field Modification #3, except as noted elsewhere in this checklist.
Health of Test Organisms via laboratory	WS#12	9. Yes. Average survival was 93.8%.
negative control:	WS# 28	10. No. The average ash free dry weigh in
 9. Control survival: ≥ 70% 10. Average ash free dry weight: ≥ 0.48 mg per surviving individual 		the control treatment was 0.425 mg. Test treatment growth ranged from 0.516 – 0.731.
Health of Test Organisms via laboratory positive control (reference toxicant):	WS#12 WS# 28 MOD#3	11. Yes. A 48-hour KCl reference toxicant test was conducted.
11. A standard reference toxicity test will be conducted.	MOD#3	12. Yes. The health of organisms based on the laboratory positive control is
12. The LC50 for a positive control test should be within the mean LC50 ±2 standard deviations of the control chart.		acceptable. The 48-hour KCl toxicant test LC50 (6830.2 ppm) was within the laboratory historical control chart limits.
Acceptability of test conditions:	WS#12	13. Yes. Dissolved oxygen (DO) was
 13. Dissolved oxygen: ≥ 2.5 mg/L 14. Temperature (daily mean): 23°C ±1°C. No value exceeding limits of 23°C ±3 °C of the mean. Temperature (instantaneous): 	WS# 28 MOD#3 SOP QA- 1407 Rev. 12c	>3.3 mg/L throughout the test. 14. Yes. During the test, temperatures ranged from 21.6 – 24.0°C and the daily mean was always 23°C ±1°C. No value exceeded of 23°C ±3 °C.
23°C±3°C		15. Possibly Not Acceptable.
15. Alkalinity, Hardness, and Ammonia: Should not vary by more than 50% during the test		Alkalinity and ammonia differences between test initiation and termination were acceptable.
16. Monitoring Requirements:		Between test initiation and termination,
 Water Quality Parameter: Dissolved oxygen, pH, conductivity, and temperature. Frequency: Monitor overlying water for each treatment daily in one surrogate test vessel for each treatment prior to renewal. Water Quality Parameter: Temperature Frequency: Monitor hourly in separate test vessel. 		hardness in two treatments (LPRT11A and LPRT11D) dropped by more than 50% (190 to 60 mg/L and 140 to 60 mg/L, respectively). In general, this drop in hardness is unusual. Two potential explanations are (1) a titration or calculation error in the hardness measurement or (2) an error in the preparation of reconstituted water. Changes in hardness will impact the bioavailability of metals to the organisms. ³

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³ Personal communication (June 2010). Mick DeGraeve and Dennis McCauley, Great Lakes Environmental Center, Traverse City, MI 49686.

Attachment 1

Lower Passaic River Restoration Project Chironomus dilutus 10-Day Survival and Growth Toxicity Test SOP QA-1407 Rev. 12c

Data Quality Element	References	Verification Assessment
 Water Quality Parameter: Alkalinity, hardness, and ammonia. Frequency: Analyze in overlying water in one surrogate test vessel for each treatment at the start and end of testing Water Quality Parameter: pore water ammonia and pH Frequency: At the end of test in each sample treatment. Porewater will be from surrogate test chamber. 		16. Yes. Water quality conditions during the test were monitored at the frequency specified in the QAPP and SOP with the exception that it was not possible to determine from the raw data if porewater ammonia and pH were measured in each test chamber at the end of the test.
Test conditions: 17. Eight replicates with 10 larvae/replicate chamber 18. Test organisms 2 nd to 3 rd instar with 50% of organisms at 3 rd instar stage. 19. Feed daily during test	SOP QA- 1407 Rev. 12c	 17. Yes. 18. Yes. The report narrative states that at test start the organisms were 2nd and 3rd instar; 12-14 days old. 19. Yes, as stated in the report narrative.
19. Teed daily during test		It should be noted that the SOP and raw data indicate that 225mL of overlying water be added to each test chamber but the report narrative states that 175 mL of overlying water was added to each chamber.
Sample Handing 20. Preservation: ≤ 4 degrees Celsius 21. Holding Time: ≤8 weeks, preferably ≤14 Days	WS#19 MOD#3	20. Cannot be determined. According to the report narrative, sediments were collected on October 27 and 28, 2009. They were received on ice at AAT on October 30, 2009. The temperature of the sediments upon receipt was not
22. All toxicity testing will be performed using the same two gallons of unsieved sediment.23. Samples will not be sieved prior to testing.		provided in the report. 21. Yes. Sample testing began 31 days after sample collection. Note that the report narrative states in two different sentences that testing began on October 30, 2009 and November 24, 2009. According to the raw data, testing
24. Project sediments will be stored at 2-4°C and will not be purged with inert gas once opened.		began on November 27, 2009. 22. Cannot be determined. The report narrative does not state that all toxicity testing was conducted using the same sediment samples (i.e., both <i>Ampelisca</i> and <i>Chironomus</i>). However, the custody forms identified that samples

Battelle July 7, 2010

Attachment 1 Lower Passaic River Restoration Project

Chironomus dilutus 10-Day Survival and Growth Toxicity Test SOP QA-1407 Rev. 12c

Data Quality Element	References	Verification Assessment
Data Quanty Element	References	were to be used for testing both species. 23. Yes. The raw data sheets indicate that sediment was not sieved prior to use. 24. Yes. Upon receipt the samples were refrigerated until testing was initiated on November 27, 2009. Comment on sampling traceability: Five sediment samples were tested
		(LPRT11A, LPRT11C, LPRT11D, LPRT11E, and LPRT16A). Accutest chain of custody forms were included in the data package for five soil samples (09910, 09911, 09912, 09913, and 09914). Based on the report package, there is no mechanism to match the custody form sample identification numbers to the reported sample values. No custody forms were provided for the test organisms or freshwater.
Delivery 25. Data turn-around time: 90 days (60 for testing and 30 for validation)	WS#30	25. Not assessed. The data report is not dated.
Validation 26. Toxicity testing data will not require full data validation. Toxicity data will only be reviewed against the acceptance limits provided in Worksheets 12 and 28.	WS#36	26. Yes. Completed as specified.
Usability 27. Usability of toxicity data is based on achieving sample holding times, acceptable water quality conditions during testing, and laboratory control treatment survival and growth criteria.	WS#37	27. Usable with reservations. Holding times, positive and negative control treatment survival, and all water quality criteria except hardness in two samples met QAPP criteria. The ash free dry weight for the negative control and the degree of change in hardness between test initiation and termination in two samples were not acceptable.

Hyalella azteca Estuarine Toxicity Test for the Lower Passaic River Restoration Project

1.0 INTRODUCTION

During October 2009, sediment samples were collected at locations along the Passaic River as part of a Remedial Investigation/Feasibility Study (RI/FS) pursuant to the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA) and the Superfund Amendment and Reauthorization Act, as agreed to by the U.S. Environmental Protection Agency (EPA) and a group of 73 companies, the Cooperative Parties Group (CPG), considered potentially responsible for contamination in the lower Passaic River. On behalf of the U.S. Army Corps of Engineers (USACE) and U.S. EPA, Malcolm Pirnie, Inc. and its subcontractor, The Louis Berger Group, Inc., provided oversight and collected and analyzed government split samples. Government split sample data will be compared to the parent samples collected by the CPG to determine if a bias exists in the data produced by the CPG.

Sediment split sample toxicity testing was performed by American Aquatic Testing, Inc. (AAT) according to the *Oversight Quality Assurance Project Plan (QAPP) for Biological Sampling, Community Surveys, and Toxicity and Bioaccumulation Testing* dated August 6, 2009 and Field Modifications No. 2 (October 15, 2009) and No. 3 (December 23, 2009). Five estuarine *Hyalella azteca* 28-day solid phase toxicity tests, representing amphipod exposure to Passaic River sediments, were conducting by AAT for the Lower Passaic River Restoration Project.

2.0 VERIFICATION PROCEDURES

An independent verification of the *Hyalella toxicity* test conditions and results was conducted by Battelle to verify that the test was conducted according to the QAPP and that the test results were acceptable. Acceptability of the toxicity test was assessed by comparing the AAT test procedures and conditions vs. the project requirements. Test procedures and results were described in the AAT report *Lower Passaic River Estuarine Section Restoration Project Sediment Toxicity Testing - Hyalella azteca* (undated). The project requirements for the toxicity tests were defined in the following project control documents:

- Oversight Quality Assurance Project Plan (QAPP) for Biological Sampling, Community Surveys, and Toxicity and Bioaccumulation Testing (August 6, 2009)
- *QAPP* Field Modifications No. 2 (October 15, 2009)
- *QAPP* Field Modification No. 3 (December 23, 2009)
- Assessment Toxicity (28-Day) of Sediments to the Amphipod, *Hyalella azteca* based on Survival and Growth – Project Specific Document (EnviroSystems, Inc. SOP QA-1467 Rev 7g)

Toxicity test verification was initiated by identifying the test requirements defined in the above documents. In particular, the QAPP Worksheets (WSs) #36 and 37 define the acceptance criteria as those contained in WSs #12 and #28. In addition, The Louis Berger Group, Inc. statement of work indicated that tests should be verified vs. the QAPP, field modifications, revised toxicity SOPs, and issues encountered. The test requirements were tabulated in a checklist (Attachment 1), which was used to guide the review.

Hyalella azteca Estuarine Toxicity Test for the Lower Passaic River Restoration Project

3.0 VERIFICATION RESULTS

According to the QAPP, toxicity test acceptability is based on the health of the organisms and the acceptability of test conditions (WSs 12 and 28). The verification of these criteria is summarized below. The checklist provided as Attachment 1 details the full test verification results.

1. Health of Organisms (Laboratory negative control)

The health of organisms based on the laboratory negative control is verified as **acceptable**. Average negative control survival was 91.3% vs. the QAPP requirement of $\geq 80\%$.

The health of organisms based on average dry weight of surviving organisms is determined to be **acceptable.** The control treatment average dry weight was 0.427 mg vs. the QAPP requirement of ≥ 0.15 mg per surviving individual.

2. Health of Organisms (Laboratory positive control):

The health of organisms based on the laboratory positive control **cannot be determined**. The reference toxicant test was run for 48-hours with KCl rather than 96-hours with cadmium chloride as specified in SOP QA-1667 Rev. 7g. The 48-hour KCl reference toxicant test LC50 (408.1 ppm) was within the laboratory historical control chart limits. However, the statistics report provided in the report package for this test does not match the report narrative results. The correct test results should be provided.

3. Acceptability of test conditions

The test conditions during the test are verified as **acceptable**, with the exception of alkalinity and hardness which are **Possibly Not Acceptable**, and the absence of salinity data.

- Dissolved oxygen concentrations were >4.5 mg/L throughout the test and are **acceptable**. The QAPP states that dissolved oxygen concentrations must be ≥ 2.5 mg/L throughout the test.
- Temperatures of overlying water ranged from 21.3 24.0°C throughout the test and are **acceptable**. The QAPP states that daily mean temperature must be within 23°C ±1°C, no temperature value may exceed 23°C ±3 °C of the mean at any time, and the instantaneous temperature must always 23°C±3°C. All QAPP criteria were achieved with two minor exceptions: on November 4 and 5, 2009 the daily mean was 21.9 °C and 21.8°C, respectively. These very minor excursions from the requirement have no impact on the test.
- Alkalinity concentration differences between test initiation and termination ranged between 10 and 67%. The QAPP states that alkalinity concentrations should not vary by more than 50% during the test. Between test initiation and termination, alkalinity in two test treatments (LPRT01F and LPRT01G) dropped by more than 50% (67 and 61%, respectively). The alkalinity conditions for these two tests are unacceptable.
- Hardness concentration differences between test initiation and termination ranged between 0.4 and 67%. The QAPP states that hardness concentrations should not vary

Hyalella azteca Estuarine Toxicity Test for the Lower Passaic River Restoration Project

by more than 50% during the test. Between test initiation and termination, hardness in two test treatments (LPRT02F and LPRT03A) dropped by more than 50% (61% and 67%, respectively). The hardness conditions for these two tests are **unacceptable.** As discussed in the checklist (Attachment 1), this drop in hardness is unusual and should be further examined by the testing laboratory. Changes in hardness will impact the bioavailability of metals to the organisms.

- Ammonia concentration differences at test initiation ranged from 0.0 to 0.05 mg/L and at test termination ranged from 0.01 to 0.13 mg/L. The QAPP states that ammonia concentrations should not vary by more than 50% during the test. However, these values were too low to calculate meaningful percent differences. At these low levels, ammonia concentrations were **acceptable**.
- Water quality conditions during the test were monitored at the frequency specified in the QAPP and SOP with the exception that salinity was not measured during the test as specified in the QAPP and SOP and that the total organic content of the sediments was not measured in a surrogate container at the start of the test. Water quality monitoring is judged to be acceptable, however, the laboratory should calculate and provide salinity values for all test treatments using the measured conductivity data.

4.0 ASSESSMENT OF USABILITY

The *Hyalella azteca* estuarine test results are verified as **acceptable with reservations**. Holding times, negative control treatment survival, dry weight results, and all water quality criteria except alkalinity and hardness met QAPP criteria. The positive control was run for 48 hours with KCl and was within laboratory control limits but the SOP specified that the positive control be a 96 hour CdCl test. For samples LPRT02F and LPRT03A, hardness dropped more than 50% between test initiation and termination and was not acceptable. For samples LPRT01F and LPRT01G, alkalinity dropped more than 50% between test initiation and termination and was not acceptable. No salinity data were reported for this test. Attachment 1 provides a full assessment of the toxicity test procedures and results vs. the QAPP requirements.

Attachment 1 **Lower Passaic River Restoration Project** Hyalella azteca Estuarine 28-Day Survival and Growth Toxicity Test **SOP QA-1467 Rev. 7g**

Data Quality Element	References	Verification Assessment
 Test Design Test approximately five sediments that are estuarine (≥5 ppt salinity) using the <i>Hyalella azteca</i> 28-day survival and growth toxicity test. Testing will follow EnviroSystems SOP QA-1467 Rev. 7g <i>H. azteca</i> organisms for testing will be purchased from the same supplier used by EnviroSystems (ARO, Inc. Hampton , NH). <i>H. azteca</i> organisms will include individuals acclimated to 10 ppt salinity. Artificial substrate for controls will be supplied by ARO and used to conduct one control sample test. Seawater will be supplied by ARO and filtered, 100 μm, prior to dilution. Sediment samples will not be sieved prior to testing. 	WS#10 WS#11 WS#18 WS#19 WS#23 MOD#3 ¹ SOP QA- 1467 Rev. 7g	 Yes, as modified by Field Modification #3. The report narrative states that samples with a porewater salinity of ≥ 5 ppt were tested using overlying water with a salinity of 10 ppt. Yes, as modified by Field Modification #3. Note: an additional, initial overlying water replacement that was not described in the SOP was conducted. 24 hours after sediment and overlying water was added to the test chambers, the overlying water was removed and new fresh water was added to the sediment. The additional water replacement does not impact the test results because overlying water is renewed twice daily throughout the test. Yes. The report narrative states that test organisms were supplied by ARO, cultured at 10 ppt, and were held under test conditions prior to testing. Yes. The report narrative states that the control sample was tested using artificial sediment provided by EnviroSystems. Yes. The report narrative states that overlying water used for exposure was created using natural salt water (26 ppt) provided by EnviroSystems and deionized water to adjust water to the exposure level of 10 ppt. The report narrative and raw data do not indicate that the seawater was filtered by ARO. Yes. The report narrative and raw data indicate that sediment was not sieved.
7. The results of the toxicity test will be statistically compared to comparable tests conducted with control sediment for control survival and/or growth.	WS#11	7. Yes. Significance vs. the control test was determined using ANOVA and Dunnett's pairwise comparisons.
8. Toxicity tests will be conducted according to the government assigned lab SOPs, modified so that	WS#11 MOD#3	8. Yes, as modified by Field Modification #3 except as noted elsewhere in this

¹ Field Modification #3 specifies that the changes to Worksheet #23 defined in the modification are applicable to split sample toxicity testing conducted after November 11, 2009. Ampelisca abdita toxicity testing was initiated on November 5, 2009, however, several of the modifications were discussed during a meeting conducted on October 21, 2009.

Lower Passaic River Restoration Project Hyalella azteca Estuarine 28-Day Survival and Growth Toxicity Test SOP QA-1467 Rev. 7g

Data Quality Element	References	Verification Assessment
test conditions are comparable to the CPG assigned laboratory SOP.		checklist.
 Health of Test Organisms via laboratory negative control: 9. Control survival: ≥ 80% 10. Average dry weight: ≥ 0.15 mg per surviving individual 	WS#12 WS# 28 SOP QA- 1467 Rev. 7g	9. Yes. Average survival was 91.3%.10. Yes. The average dry weigh in the control treatment was 0.427 mg.
 Health of Test Organisms via laboratory positive control (reference toxicant): 11. A 96-hour water only standard reference toxicity test will be conducted with cadmium chloride 12. A separate reference toxicant test will be conducted for estuarine organisms. 13. The LC50 for a positive control test should be within the mean LC50 ±2 standard deviations of the control chart. 	WS#12 WS# 28 MOD#3 SOP QA- 1467 Rev. 7g	 Cannot be determined. The reference toxicant test was run for 48-hours with KCl rather than 96-hours with cadmium chloride. Cannot be determined. Salinity was not measured in the reference toxicant test. Initial conductivity ranged from 15590 μmhos in the controls to 18410 μmhos in the 2000 ppm exposure. Yes. The narrative reports that the LC50 for the 48-hour KCl reference toxicant test was 408.1 ppm and that this value fell within the control chart limits. However, the statistics report provided in the report package for this test does not match the report narrative results. The correct test results should be provided.
 Acceptability of test conditions: 14. Overlying water quality (i.e., freshwater vs. saline water) will be consistent with exposures conducted by EnviroSystems, Inc. 15. Dissolved oxygen: ≥ 2.5 mg/L 16. Temperature (daily mean): 23°C ±1°C. No value exceeding limits of 23°C ±3 °C of the mean. Temperature (instantaneous): 23°C±3°C 17. Alkalinity, Hardness, and Ammonia: Should not vary by more than 50% during the test 18. Monitoring Requirements: • Water Quality Parameter: Dissolved 	WS#12 WS# 28 MOD#3 SOP QA- 1467 Rev. 7g	 14. Cannot be determined. Conditions for EnviroSystems tests were not available for comparison. This assessment will be performed when ATT and EnviroSystems data are compared. 15. Yes. Dissolved oxygen (DO) was >4.5 mg/L throughout the test. 16. Yes. During the test, temperatures ranged from 21.3 – 24.0°C and the daily mean was always 23°C ±1°C with two minor exceptions: on November 4 and 5, 2009 the daily mean was 21.9 °C and 21.8°C, respectively. These very minor excursions from the requirement have no impact on the test. No value exceeded of 23°C ±3 °C. 17. Possibly Not Acceptable.
oxygen, pH, specific conductance, salinity, and temperature.		Possibly Not Acceptable. Between test initiation and termination,

Lower Passaic River Restoration Project Hyalella azteca Estuarine 28-Day Survival and Growth Toxicity Test SOP QA-1467 Rev. 7g

Data Quality Element	References	Verification Assessment
 Frequency: Monitor overlying water for each treatment daily in one surrogate test vessel for each treatment prior to renewal. Water Quality Parameter: Temperature Frequency: Monitor hourly in separate test vessel. Water Quality Parameter: Conductivity Frequency: daily prior to use in assay. Water Quality Parameter: Alkalinity, hardness, and ammonia. Frequency: Analyze in a surrogate test vessel for each treatment at test start and weekly thereafter. Water Quality Parameter: Total organic carbon content (measured as loss on ignition) Frequency: Measure in surrogate container at test start and end. 	TWO CHECK	alkalinity in two test treatments (LPRT01F and LPRT01G) dropped by more than 50% (67 and 61%, respectively). • Between test initiation and termination, hardness in two test treatments (LPRT02F and LPRT03A) dropped by more than 50% (61 and 67%, respectively). In general, drops in hardness are unusual. Two potential explanations are (1) a titration or calculation error in the hardness measurement or (2) an error in the preparation of reconstituted water. Changes in hardness will impact the bioavailability of metals to the organisms. ² • Ammonia concentrations at test initiation ranged from 0.0 to 0.05 mg/L and at test termination ranged from 0.01 to 0.13 mg/L. At these low levels, ammonia concentrations were acceptable regardless of the calculated percent difference. Note that in most cases, the percent difference cannot be calculated because the ammonia concentration was 0.0 mg/L.
		18. Yes. Water quality conditions during the test were monitored at the frequency specified in the QAPP and SOP with two exceptions:
		 No salinity data were measured or calculated although conductivity was measured. No criteria are defined for conductivity in the QAPP or SOP, but the estuarine test salinity was defined as 10 ppt. Salinity data should be calculated and reported for all test treatments.
		 Total organic content of the sediments was not measured in a

² Personal communication (June 2010). Mick DeGraeve and Dennis McCauley, Great Lakes Environmental Center, Traverse City, MI 49686.

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Lower Passaic River Restoration Project Hyalella azteca Estuarine 28-Day Survival and Growth Toxicity Test SOP QA-1467 Rev. 7g

Data Quality Element	References	Verification Assessment
		surrogate container at test start.
 Special considerations for the impact of estuarine conditions on Hyalella azteca toxicity data from WS#23 Footnote 2: 19. Salinity in porewater will be measured prior to testing. Samples having a porewater salinity of ≤5 ppt will be tested using freshwater as the overlying water. Samples with porewater salinity > 5 ppt will be tested using 10 ppt salinity overlying water. Due to concern regarding the usability of Hyalella azteca toxicity data from the estuarine section of the river where salinity levels are >15 ppt, the interstitial salinity in the sediment samples will be measured in the laboratory, and the interstitial salinity > 8 ppt will be adjusted to a range of 5 to 7 ppt before test initiations. The adjustment will be performed by replacing the overlaying freshwater in each beaker (the sediments will not be manually mixed with fresh water) and incorporating a salinity control into the test design. 20. The Hyalella toxicity test results from estuarine sampling areas will be evaluated by comparing the survival and growth results of the negative control with a salinity-adjusted control (the negative control sediment for the Ampelisca toxicity test). Salinity Control survival (compared to 	WS#20 WS#23 SOP QA- 1467 Rev. 7g	
 survival of negative control for <i>Ampelisca abdita</i> toxicity test). Salinity Control growth (compared to growth of negative control for <i>Ampelisca abdita</i> toxicity test). 		
Test conditions:	SOP QA-	21. Cannot be determined. The report
21. Parent <i>Hyalella</i> culture will be	1467 Rev. 7g MOD#2	narrative states that test organisms were received from ARO and

Lower Passaic River Restoration Project Hyalella azteca Estuarine 28-Day Survival and Growth Toxicity Test SOP QA-1467 Rev. 7g

Data Quality Element	References	Verification Assessment
acclimated to 10 ppt salinity by the CPG to generate successive daughter individuals for testing. 22. Test organisms will be selected from cultures of appropriate salinity (freshwater, <0.5ppt, or 10 ppt) depending on the porewater salinity of an individual sample. 23. Eight replicates with 10 larvae/replicate chamber 24. Test organisms 7-8 days old. 25. Feed daily during test		acclimated at AAT but the parent history was not provided. 22. Cannot be determined. It is not possible to determine if test organisms were selected from appropriate salinity hatches. The narrative states that test organisms were acclimated to the SOP-specified water quality conditions prior to testing but the salinity of water in which organisms were hatched was not provided. 23. Yes. 24. Yes. The report narrative states that the test organisms were 7-8 days old. 25. Yes, the raw data directs, and the report narrative states, that organisms were
Sample Handing 26. Preservation: ≤ 4 degrees Celsius 27. Holding Time: ≤8 weeks, preferably ≤14 Days	WS#19 MOD#3	fed daily. 26. Cannot be determined. According to the report narrative, sediments were collected on October 13 and 14, 2009. They were received on ice at AAT on October 16, 2009. The temperature of the sediments upon receipt was not provided in the report.
28. All toxicity testing will be performed using the same two gallons of unsieved sediment.		27. Yes. Sample testing began on November 4, 2009, 22 days after sample collection.
 29. Samples will not be sieved prior to testing. 30. Project sediments will be stored at 2-4°C and will not be purged with inert gas once opened. 		 28. Cannot be determined. The report narrative does not state that all toxicity testing was conducted using the same sediment samples (i.e., both <i>Hyalella</i> and <i>Ampelisca</i>). However, the custody forms identified that samples were to be used for testing both species. 29. Yes. The raw data sheets indicate that sediment was not sieved prior to use. 30. Yes. Upon receipt the samples were refrigerated until testing was initiated on November 4, 2009.
		Comment on sample traceability: Five sediment samples were tested (LPRT02F, LPRT03A, LPRT01F,

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Data Quality Element	References	Verification Assessment
		LPRT02A, and LPRT01G). The report package did not include the custody forms for these samples. Accutest chain of custody forms were included in the data package for three AQ samples (09839, 09841, and 09842) and two soil samples (09843 and 09844). Based on the report package, there is no mechanism to match the custody form sample identification numbers to the reported sample values. No custody forms were provided for the test
Delivery 31. Data turn-around time: 90 days (60 for testing and 30 for validation)	WS#30	organisms. 31. Not assessed. The data report is not dated.
Validation 32. Toxicity testing data will not require full data validation. Toxicity data will only be reviewed against the acceptance limits provided in Worksheets 12 and 28.	WS#36	32. Yes. Completed as specified.
Usability 33. Usability of toxicity data is based on achieving sample holding times, acceptable water quality conditions during testing, and laboratory control treatment survival and growth criteria (sic).	WS#37	33. Usable with reservations. Holding times, control treatment survival, dry weights, and all water quality criteria except alkalinity and hardness met QAPP criteria.

Hyalella azteca Freshwater Toxicity Test for the Lower Passaic River Restoration Project

1.0 INTRODUCTION

During October 2009, sediment samples were collected at locations along the Passaic River as part of a Remedial Investigation/Feasibility Study (RI/FS) pursuant to the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA) and the Superfund Amendment and Reauthorization Act, as agreed to by the U.S. Environmental Protection Agency (EPA) and a group of 73 companies, the Cooperative Parties Group (CPG), considered potentially responsible for contamination in the lower Passaic River. On behalf of the U.S. Army Corps of Engineers (USACE) and U.S. EPA, Malcolm Pirnie, Inc. and its subcontractor, The Louis Berger Group, Inc., provided oversight and collected and analyzed government split samples. Government split sample data will be compared to the parent samples collected by the CPG to determine if a bias exists in the data produced by the CPG.

Sediment split sample toxicity testing was performed by American Aquatic Testing, Inc. (AAT) according to the *Oversight Quality Assurance Project Plan (QAPP) for Biological Sampling, Community Surveys, and Toxicity and Bioaccumulation Testing* dated August 6, 2009 and Field Modifications No. 2 (October 15, 2009) and No. 3 (December 23, 2009). Five freshwater *Hyalella azteca* 28-day solid phase toxicity tests, representing amphipod exposure to Passaic River sediments, were conducting by AAT for the Lower Passaic River Restoration Project.

2.0 VERIFICATION PROCEDURES

An independent verification of the *Hyalella toxicity* test conditions and results was conducted by Battelle to verify that the test was conducted according to the QAPP and that the test results were acceptable. Acceptability of the toxicity test was assessed by comparing the AAT test procedures and conditions vs. the project requirements. Test procedures and results were described in the AAT report *Lower Passaic River Freshwater Section Restoration Project Sediment Toxicity Testing - Hyalella azteca* (undated). The project requirements for the toxicity tests were defined in the following project control documents:

- Oversight Quality Assurance Project Plan (QAPP) for Biological Sampling, Community Surveys, and Toxicity and Bioaccumulation Testing (August 6, 2009)
- *OAPP* Field Modifications No. 2 (October 15, 2009)
- *QAPP* Field Modification No. 3 (December 23, 2009)
- Assessment Toxicity (28-Day) of Sediments to the Amphipod, *Hyalella azteca* based on Survival and Growth – Project Specific Document (EnviroSystems, Inc. SOP QA-1467 Rev. 7g)

Toxicity test verification was initiated by identifying the test requirements defined in the above documents. In particular, the QAPP Worksheets (WSs) #36 and 37 define the acceptance criteria as those contained in WSs #12 and #28. In addition, The Louis Berger Group, Inc. statement of work indicated that tests should be verified vs. the OAPP, field modifications, revised toxicity SOPs, and

Hyalella azteca Freshwater Toxicity Test for the Lower Passaic River Restoration Project

issues encountered. The test requirements were tabulated in a checklist (Attachment 1), which was used to guide the review.

3.0 VERIFICATION RESULTS

According to the QAPP, toxicity test acceptability is based on the health of the organisms and the acceptability of test conditions (WSs 12 and 28). The verification of these criteria is summarized below. The checklist provided as Attachment 1 details the full test verification results.

1. Health of Organisms (Laboratory negative control)

The health of organisms based on the laboratory negative control is verified as **acceptable**. Average negative control survival was 97.5% vs. the QAPP requirement of $\geq 80\%$.

The health of organisms based on average dry weight of surviving organisms is determined to be **acceptable.** The control treatment average dry weight was 0.427 mg vs. the QAPP requirement of ≥ 0.15 mg per surviving individual. However, dry weights were only determined for organisms from samples with acceptable survival. This is a deviation from the SOP which states "all surviving amphipods from an individual replicate are ... dried and ... weighed to the nearest 0.01 mg."

2. Health of Organisms (Laboratory positive control):

The health of organisms based on the laboratory positive control **cannot be determined**. The reference toxicant test was run for 48-hours with KCl rather than 96-hours with cadmium chloride as specified in SOP QA-1667 Rev. 7g. The 48-hour KCl toxicant test LC50 (395.3 ppm) was within the laboratory historical control chart limits.

3. Acceptability of test conditions

The test conditions during the test are verified as **acceptable**, with the exception of alkalinity and hardness which are **Possibly Not Acceptable**, and the absence of salinity data.

- Dissolved oxygen concentrations were >4.1 mg/L throughout the test and are **acceptable**. The QAPP states that dissolved oxygen concentrations must be ≥ 2.5 mg/L throughout the test.
- Temperatures of overlying water ranged from 20.5 24.7°C throughout the test and are **acceptable**. The QAPP states that daily mean temperature must be within 23°C ±1°C, no temperature value may exceed 23°C ±3 °C of the mean at any time, and the instantaneous temperature must always 23°C±3°C. All QAPP criteria were achieved.
- Alkalinity concentration differences between test initiation and termination ranged between 14 and 40% and are **acceptable**. The QAPP states that alkalinity concentrations should not vary by more than 50% during the test. All QAPP criteria were achieved.
- Hardness concentration differences between test initiation and termination ranged between 0.0 [a questionable value] and 58% and are **Possibly Not Acceptable**. The QAPP states that hardness concentrations should not vary by more than 50% during the test. In one treatment (LPRT11A) hardness dropped by more than 50% (58%) and is **unacceptable**.

Hyalella azteca Freshwater Toxicity Test for the Lower Passaic River Restoration Project

As discussed in the checklist (Attachment 1), this drop in hardness is unusual and should be further examined by the testing laboratory. Changes in hardness will impact the bioavailability of metals to the organisms. Further, both the initial and final hardness values for Sample LPRT11C were recorded as 110 mg/L. Because the *Chironomus* test with this sample registered a hardness drop of 36% it appears that the final hardness value for this *Hyalella* treatment is a recording error.

- Ammonia concentration differences at test initiation ranged from 0.0 to 2.1 mg/L and at test termination ranged from 0.0 to 0.08 mg/L. The QAPP states that ammonia concentrations should not vary by more than 50% during the test. However, these values were too low to calculate meaningful percent differences. At these low levels, ammonia concentrations were **acceptable**.
- Water quality conditions during the test were monitored at the frequency specified in the QAPP and SOP with the exception that salinity was not measured during the test as specified in the QAPP and SOP and that the total organic content of the sediments was not measured in a surrogate container at the start of the test. Water quality monitoring is judged to be acceptable, however, the laboratory should calculate and provide salinity values for all test treatments using the measured conductivity data.

4.0 ASSESSMENT OF USABILITY

The *Hyalella azteca* freshwater test results are verified as **acceptable with reservations**. Holding times, negative control treatment survival, dry weight results, and all water quality criteria except hardness met QAPP criteria. The positive control was run for 48 hours with KCl and was within laboratory control limits but the SOP specified that the positive control be a 96 hour CdCl test. For sample LPRT11A, hardness dropped more than 50% between test initiation and termination and was not acceptable. No salinity data were reported for this test. Attachment 1 provides a full assessment of the toxicity test procedures and results vs. the QAPP requirements.

Attachment 1 Lower Passaic River Restoration Project Hyalella azteca Freshwater 28-Day Survival and Growth Toxicity Test SOP QA-1467 Rev 7g

	Data Quality Element	References		Verification Assessment
1. 2. 3. 4. 5. 6.	Test approximately five sediments that are freshwater <5 ppt salinity) using the <i>Hyalella azteca</i> 28-day survival and growth toxicity test. Testing will follow EnviroSystems SOP QA-1467 Rev. 7g H. azteca organisms for testing will be purchased from the same supplier used by EnviroSystems (ARO, Inc. Hampton NH). H. azteca organisms will include individuals acclimated to freshwater. Artificial substrate for controls will be supplied by ARO and used to conduct one control sample test. Freshwater will consist of a 50:50 (by volume) mix of natural water and re-constituted hard water created by AAT using de-ionized water (this requirement was later superseded when EnviroSystems, Inc. shipped freshwater to AAT). Freshwater will be filtered 100 µm, prior to addition to reconstituted water. Sediment samples will not be sieved prior to testing.	WS#10 WS#11 WS#18 WS#19 WS#23 MOD#3 ¹ SOP QA- 1467 Rev. 7g	 3. 5. 7. 	Yes, as modified by Field Modification #3. It was not possible to verify that the sediment samples tested using <i>Hyalella azteca</i> were collected from a freshwater location because no data for the initial porewater salinity was provided in the report package. Yes, as modified by Field Modification #3. Note: an additional, initial overlying water replacement that was not described in the SOP was conducted. 24 hours after sediment and overlying water was added to the test chambers, the overlying water was removed and new fresh water was added to the sediment. The additional water replacement does not impact the test results because overlying water is renewed twice daily throughout the test. Yes. The report narrative states that test organisms were supplied by ARO and were held under test conditions prior to testing. Yes. The report narrative states that the control sample was tested using artificial sediment provided by EnviroSystems. Yes. The report narrative states that overlying water used for exposure was created using natural freshwater provided by EnviroSystems and reconstituted fresh water prepared by AAT. The report narrative and raw data do not indicate that the freshwater was filtered. Yes. The report narrative and raw data indicate that sediment was not sieved. Yes. Significance vs. the control test was determined using ANOVA and Dunnett's determined using ANOVA and Dunnett's
	comparable tests conducted with control sediment for control survival and/or growth.			determined using ANOVA and Dunnett's pairwise comparisons.
8.	Toxicity tests will be conducted according to the government	WS#11 MOD#3	8.	Yes, as modified by Field Modification #3 and except as noted elsewhere in this

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¹ Field Modification #3 specifies that the changes to Worksheet #23 defined in the modification are applicable to split sample toxicity testing conducted after November 11, 2009. *Ampelisca abdita* toxicity testing was initiated on November 5, 2009, however, several of the modifications were discussed during a meeting conducted on October 21, 2009.

Lower Passaic River Restoration Project Hyalella azteca Freshwater 28-Day Survival and Growth Toxicity Test SOP QA-1467 Rev 7g

Data Quality Element	References	Verification Assessment
assigned lab SOPs, modified so that test conditions are comparable to the CPG assigned laboratory SOP.		checklist.
 Health of Test Organisms via laboratory negative control: 9. Control survival: ≥ 80% 10. Average dry weight: ≥ 0.15 mg per surviving individual 	WS#12 WS# 28 SOP QA- 1467 Rev. 7g	9. Yes. Average survival was 97.5%. 10. Yes. The average dry weigh in the control treatment was 0.427 mg. However, dry weights were only determined for organisms from samples with acceptable survival. This is a deviation from the SOP which states "all surviving amphipods from an individual replicate are dried and weighed to the nearest 0.01 mg."
Health of Test Organisms via laboratory positive control (reference toxicant): 11. A 96-hour water only standard reference toxicity test will be conducted with cadmium chloride (CdCl) 12. A separate reference toxicant test will be conducted for fresh water organisms. 13. The LC50 for a positive control test should be within the mean LC50 ±2 standard deviations of the control chart.	WS#12 WS# 28 MOD#3 SOP QA- 1467 Rev. 7g	 11. Cannot be determined. The reference toxicant test was run for 48-hours with potassium chloride (KCl) rather than 96-hours with cadmium chloride. 12. Cannot be determined. Salinity was not measured in the reference toxicant test. Initial conductivity ranged from 276 μmhos in the controls to 3838 μmhos in the 2000 ppm exposure. 13. Yes. The LC50 for the 48-hour KCl reference toxicant test was 395.3 ppm. This value fell within the control chart limits.
Acceptability of test conditions: 14. Overlying water quality (i.e., freshwater vs. saline water) will be consistent with exposures conducted by EnviroSystems, Inc. 15. Dissolved oxygen: ≥ 2.5 mg/L 16. Temperature (daily mean): 23°C ±1°C. No value exceeding limits of 23°C ±3 °C of the mean. Temperature (instantaneous): 23°C±3°C 17. Alkalinity, Hardness, and Ammonia: Should not vary by more than 50% during the test 18. Monitoring Requirements:	WS#12 WS# 28 MOD#3 SOP QA- 1467 Rev. 7g	 14. Cannot be determined. Conditions for EnviroSystems tests were not available for comparison. This assessment will be performed when ATT and EnviroSystems data are compared. 15. Yes. Dissolved oxygen (DO) was >4.1 mg/L throughout the test. 16. Yes. During the test, temperatures ranged from 20.5 – 24.7°C and the daily mean was always 23°C ±1°C. No value exceeded of 23°C ±3 °C. 17. Possibly Not Acceptable. Alkalinity concentration differences between test initiation and termination ranged between 14 and 40% and are
 Water Quality Parameter: Dissolved oxygen, pH, specific conductance, 		acceptable. The QAPP states that alkalinity concentrations should not

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	Data Quality Element	References	Verification Assessment
•	salinity, and temperature. Frequency: Monitor overlying water for each treatment daily in one surrogate test vessel for each treatment prior to renewal. Water Quality Parameter: Temperature Frequency: Monitor hourly in separate test vessel. Water Quality Parameter: Conductivity Frequency: daily prior to use in assay. Water Quality Parameter: Alkalinity, hardness, and ammonia. Frequency: Analyze in a surrogate test vessel for each treatment at test start and weekly thereafter.	References	vary by more than 50% during the test. All QAPP criteria were achieved. • Hardness concentration differences between test initiation and termination ranged between 0.0 [a questionable value] and 58%. The QAPP states that hardness concentrations should not vary by more than 50% during the test. In one treatment (LPRT11A) hardness dropped by more than 50% (58%) and is unacceptable. It is unusual that there was no change in hardness for Sample LPRT 11C between test initiation and termination (110 mg/L) because in the <i>Chironomus</i> test for this sample the hardness dropped from 110 mg/L to 70 mg/L at test termination. In general, drops in hardness are unusual.
•	Sediment Quality Parameter: Total organic content (measured as loss on ignition) Frequency: Measure in surrogate container for each sediment.		Two potential explanations are (1) a titration or calculation error in the hardness measurement or (2) an error in the preparation of reconstituted water. Changes in hardness will impact the bioavailability of metals to the organisms. ² • Ammonia concentration differences at test initiation ranged from 0.0 to 2.1 mg/L and at test termination ranged from 0.0 to 0.08 mg/L. The QAPP states that ammonia concentrations should not vary by more than 50% during the test. However, these values were too low to calculate meaningful percent differences. At these low levels, ammonia concentrations were acceptable. 18. Yes. Water quality conditions during the test were monitored at the frequency specified in the QAPP and SOP with two exceptions: • No salinity data were measured or calculated. Conductivity was measured. No criteria are defined

² Personal communication (June 2010). Mick DeGraeve and Dennis McCauley, Great Lakes Environmental Center, Traverse City, MI 49686.

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Data Quality Element	References	Verification Assessment
		for either salinity or conductivity in the QAPP or SOP.
		 Total organic content of the sediments was not measured in a surrogate container at test start.
 Test conditions: 19. Parent <i>Hyalella</i> culture will be acclimated to 10 ppt salinity by the CPG to generate successive daughter individuals for testing. 20. Test organisms will be selected from cultures of appropriate salinity (freshwater, <0.5ppt, or 10 ppt) depending on the porewater salinity of an individual sample. 21. Eight replicates with 10 larvae/replicate chamber 22. Test organisms 7-8 days old. 23. Feed daily during test 	SOP QA- 1467 Rev. 7g MOD#2	 Cannot be determined. The report narrative states that test organisms were received from ARO and acclimated at AAT but the parent history was not provided. Cannot be determined. It is not possible to determine if test organisms were selected from appropriate salinity hatches. The narrative states that test organisms were acclimated to the SOP-specified water quality conditions prior to testing but the salinity of water in which organisms were hatched was not provided. Yes. Yes. The report narrative states that the test organisms were 7-8 days old. Yes, the raw data directs, and the report narrative states, that organisms were fed daily.
 Sample Handing 24. Preservation: ≤4 degrees Celsius 25. Holding Time: ≤8 weeks, preferably ≤14 Days 26. All toxicity testing will be performed using the same two gallons of unsieved sediment. 27. Samples will not be sieved prior to testing. 28. Project sediments will be stored at 2-4°C and will not be purged with inert gas once opened.¹ 	WS#19 MOD#3	 24. Cannot be determined. According to the report narrative, sediments were collected on October 27 and 28, 2009. They were received on ice at AAT on October 30, 2009. The temperature of the sediments upon receipt was not provided in the report. (Note that the report narrative states that samples arrived on October 16th, but this does not agree with the sample custody forms). 25. Yes. Sample testing began on November 24, 2009, 28 days after sample collection. 26. Cannot be determined. The report narrative does not state that all toxicity testing was conducted using the same sediment samples (i.e., both <i>Hyalella</i> and <i>Chironomus</i>). However, the

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Data Quality Element	References	Verification Assessment
		custody forms identified that samples were to be used for testing both species. 27. Yes. The raw data sheets indicate that sediment was not sieved prior to use. 28. Yes. Upon receipt the samples were refrigerated until testing was initiated
		on November 24, 2009. Comment on sampling traceability: Five sediment samples were tested
		(LPRT11A, LPRT11C, LPRT11D, LPRT11E, and LPRT16A). Accutest chain of custody forms were included in the data package for five soil samples (09910, 09911, 09912, 09913, and 09914). Based on the report package, there is no mechanism to match the custody form sample identification numbers to the reported sample values.
		No custody forms were provided for the test organisms or freshwater. There is no dated signature on the custody forms relinquishing samples collected on October 27, 2009.
Delivery 29. Data turn-around time: 90 days (60 for testing and 30 for validation)	WS#30	29. Not assessed . The data report is not dated.
Validation 30. Toxicity testing data will not require full data validation. Toxicity data will only be reviewed against the acceptance limits provided in Worksheets 12 and 28.	WS#36	30. Yes. Completed as specified.
Usability 31. Usability of toxicity data is based on achieving sample holding times, acceptable water quality conditions during testing, and laboratory control treatment survival and growth criteria.	WS#37	31. Usable with reservations. Holding times, control treatment survival and dry weight and all water quality criteria except hardness met QAPP criteria. The positive control was run for 48 hours with KCl and was within laboratory control limits but the SOP specified that the positive control be a 96 hour CdCl test. For sample LPRT11A, hardness dropped more than 50% between test initiation and

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Data Quality Element	References	Verification Assessment
		termination and was not acceptable. Salinity was not reported for this test.